Reference Manual

FISH CULTURE MANUAL

by
FRED Staff

Alaska Department of Fish and Game
Division of Fisheries Rehabilitation,
Enhancement and Development

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June 1983

Fish Culture Manual/by FRED staff. -- Juneau, Alaska (P.O. Box 3-2000, Juneau 99802): Alaska Department of Fish and Game, Division of Fisheries Rehabilitation, Enhancement and Development, (1983).
90 p.; 28 cm.
"June 1983"
Bibliography: p. 86-90
1. Fish-culture-Alaska-Handbooks, manuals, etc. 2. Fish hatcheries--Alaska--Handbooks, manuals, etc. I. Alaska. Division of Fisheries Rehabilitation, Enhancement and Development.
SH35.A62F53 1983

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INTRODUCTION

History

Fish culture dates back to several thousand years B.C. Originally, fish were cultured only in the static-water environment of ponds. Such ponds served both as home and food producer for the fish in much the same way that uncultivated land is utilized by livestock. Later, pond fertilization increased food production resulting in increased fish production. This technique is analogous to the cultivation and fertilization of pastures for grazing.

To attain higher levels of production per unit of space, food must be delivered to the fish pond from outside. However, there are limitations to this feeding technique. As a result of metabolic activity, fish extract oxygen and discharge waste into the water. Thus, if higher production levels are desired, oxygen must be replenished and waste must be removed.

Salmonids have been reared in flow-through ponds for some 200 years (Slack 1872). The pond serves only as a home. Food is delivered from outside while a continuous supply of fresh water flows through the pond delivering oxygen and carrying away waste. This method of fish production is called "intensive" and requires less volume than the "extensive" or static-pond method. In intensive culture, the production level per unit volume is directly proportional to the amount of fresh water delivered to the pond. In other words, the higher the rate of water exchange, the higher the production.

Objectives

There are four major goals of fish production:

1) Stocking waters for recreational, subsistence, and commercial fisheries.
2) Production of food.
3) Production of bait fish.
4) Production of aquaria fish.

This manual deals only with the first goal, but some of the principles discussed are applicable to the other three as well.

Fish hatcheries must meet definite objectives. Fish must be available at the right time and be of the right size, species, stock, and number. Shortages and excesses create problems, not only to fisheries managers but also to program planners, policy makers, and administrators. Therefore, accuracy is necessary.

Hatcheries must attain the following objectives:

1) Healthy fish.
2) Target number.
3) Target size.

4) High survival.
5) Feeding efficiency.
6) Energy efficiency.
7) Labor efficiency.

These points warrant further discussion in order to provide hatchery personnel with some direction on how these objectives may be realized.

Healthy Fish:

Fish should not be reared under undue stress. Stress may be caused by overcrowding, water that is too warm or silty or that flows at too great a velocity, and disturbances such as tank cleaning, human activity, or predation. The culturist should be alert for these and other conditions that may stress the fish. This often means good, commonsensical practices in hatchery management, but it also involves early observation of declining performance, such as reduction in growth rate or a change in behavior that could reflect reduced general vitality. There is no substitute for the watchful eye of the culturist who is in daily "contact" with his fish. Indeed, good fish care requires knowledge and dedication, motivated by a genuine interest and concern for the welfare of the fish.

Egg Quality. Improper spawning techniques may yield poor quality eggs, low fertilization rates, and poor quality fry. A vigorous brood stock can be maintained through proper nutrition, genetic selection, careful handling, and a healthy environment. Eggs taken from females that are not at the peak of ripeness will often be of poor quality. The curve for viability of eggs, relative to ripeness, has a sharp peak rather than the gradual, bell-shaped curve that one might expect.

Water Quality. There is little hope for successful or efficient fish production without adequate water quality. Source water must meet the proper physical and chemical criteria for the desired species of fish; otherwise, pre-treatment is necessary. This is expensive and should be avoided if possible. However, many successful hatcheries have been built on water sources which, seasonally or occasionally, become substandard due to undesirable temperature levels, siltation, and dissolved oxygen fluctuations.

Apart from permanent corrective measures, which may involve the services of engineers and construction crews, the hatchery manager has several immediate options in the event of a sudden decrease in water quality. These include reductions in loading densities, reduction or temporary cessation of feeding, aeration of water, and increase of water flow.

Water quality will always deteriorate as loading increases. The hatchery manager must not exceed established loading levels. These are aimed at maximal efficiency while maintaining a good rearing environment for the fish. Water quality problems, when they are caused by overloading or improper flow rates, should be recognized and corrected immediately. The alert and responsible manager knows at all times what his actual fish loadings are, relative to the allowable limit.

Of all the water quality factors, probably none is more critical than water temperature. Once temperature rises beyond the optimum, conditions deteriorate rapidly. Rising temperatures, which are sometimes encountered in Alaska, have a multiplicity of effects:

1) A fish's metabolic rate increases, increasing its demand for oxygen, while the solubility of oxygen in water decreases.
2) The danger of disease increases because most fish pathogens are favored at warmer temperatures.
3) The ability of fish to resist infections diminishes (Fryer 1974).
4) Supersaturation of dissolved gasses may occur, especially when water is heated artificially.

Stress. Stress may be caused by poor water quality, but also by handling, disturbances, and light conditions. Moreover, not enough is known about exactly how much stress a fish can tolerate. Stress reaction depends upon the general condition of the fish and its environment. As a rule, it is best to minimize stress.

Automatic feeders eliminate the need for human activity along the rearing units. Dip nets, live crates, and raceway sections should not be over-loaded. Extensive handling, often required when marking fish, should only be done with healthy fish under proper conditions of temperature and dissolved oxygen. Fish should always remain wet and be returned to water as quickly as possible.

Unnecessary stress must be avoided. Although healthy fish are resilient to moderate amounts of handling, etc., the effects of stress are additive. When stressors are piled one upon the other, problems will occur. For example, handling fish for inventory or transportation when they are suffering from bacterial gill disease or furunculosis is devastating. Warm water and water low in dissolved oxygen reduce a fish's resistance to parasitic or bacterial infections such as furunculosis or vibriosis. Overfeeding young fry with dusty diets can lead to gill disease; overfeeding of rainbow trout in cold water can cause excessive fat deposits that may kill them when the water warms.

Food Quality. Good food is extremely important to the health of the fish. A decline in growth rate and an increase in food conversion can reflect food quality. A nutritionally inadequate diet can produce symptoms ranging from the obvious to the subtle. Although nutritionally sound diets are now readily available for trout and salmon, food-related problems should not be ruled out. When diets are suspected of causing poor health or increased mortalities in fish, the hatchery manager should contact his immediate supervisor, the fish pathology section, and the chief of Technology and Development.

The hatchery manager must know the shelf life of fish food and how to handle and store it. Quality control at the manufacturing plant is also very important and, ideally, should be performed on a random basis. The hatchery manager can perform some quality controls such as checking for percent dust and fines, for composition (large particles of one type of ingredient or various irregularities), for odor (rancidity), and for consistency (firmness and water stability).

Poor food quality may be due to use beyond normal shelf life, mishandling, improper manufacture leading to contamination or poor mixing, or adventitious toxins.

The primary cause of a problem may be difficult to identify when a secondary agent or cause manifests itself clearly, masking the primary one. Many disease outbreaks are, in actuality, manifestations of another primary problem.

Pathogens. Diseases often cause mortalities in hatchery fish. An estimated one-third of the cost of fish production nationwide is due to diseases (Westers 1979).

Absolute disease prevention is only possible when the disease-causing agents are not permitted to contact their hosts. Fish can never be reared economically in a truly sterile environment, but so-called disease-free hatcheries are desirable. These are hatcheries where the water source is free from specific fish pathogens that are, primarily, obligatory disease organisms whose continued existence depends upon the fish. Such water sources, therefore, must be free of fish and pathogens. The best water sources are often wells and underground springs.

Another approach is the treatment of water before it enters the hatchery. Certain larger parasites can be filtered out, but bacteria and viruses must be killed with ozone or ultraviolet radiation. These processes are costly and not without risks. They are most practical for relatively small water flows such as those that are used during incubation and fry rearing.

The presence of disease organisms, either in the water source or within the fish themselves, does not automatically result in a disease outbreak and mortality. Diseases only occur when a susceptible host and virulent pathogen meet under certain environmental conditions. For example, an overcrowded pond with water low in dissolved oxygen and high in organic debris provides an environment favorable to endemic pathogens. The same principles apply in the human environment where, in many situations, the microbe has a constant and ubiquitous presence that causes disease only when some weakening of the host by another factor allows infection to proceed. Infectious disease is composed of three variables: the host, pathogen, and environment. The fish culturist is concerned with lowering the numbers of pathogens in his hatchery and with eliminating unnecessary stress from the environment.

Target Number:

Proper management requires a specific number of fish on hand throughout each phase of the incubation and rearing cycle. Accurate mortality records must be maintained. Fish production records over a 3- to 5-year period will allow the manager to establish a production performance index by species for the hatchery. Starting a production cycle with many more eggs than necessary is poor management. Eggs, although relatively inexpensive to incubate, are costly to procure.

As the rearing cycle progresses, the cost of excess fish escalates rapidly due to an increasing demand for food, care, and space. Only with the aid of accurate hatchery management records can the most rational production program be established for each species at a particular hatchery. Until a hatchery manager has enough data to compute a survival index for his hatchery, he will use the FRED standard survival values as a guide.

Target Size:

Fish produced for management often must meet rather precise size requirements within a definite period of time. The goals must be realistic, i.e., they must be compatible with the inherent growth potential of the species. However, there are some hatchery manipulations, e.g., water temperature alteration, which can help accomplish the size and time objectives. Water temperature control can be very expensive, but it may not be necessary during the entire incubation and rearing cycle. Changing water temperature during incubation and early rearing, when water flow requirements are relatively low, may be all that is necessary.

High Survival:

The hatchery manager is responsible for ensuring maximal fish survival. This is accomplished through disease prevention, stress avoidance, maintenance of water quality, predator control, nutritionally sound diets, and other good cultural practices. When unusual mortality occurs during the incubation and rearing cycle, the cause must be identified so corrective and preventive measures can be applied.

Feeding Efficiency:

Feeding levels and frequencies should be adjusted to attain maximal food utilization. This can be accomplished by first examining available scientific literature and hatchery records and then by trial and error.

Feeding requirements at each hatchery may differ somewhat due to species, stocks, seasons, water quality, and temperature. Each hatchery must collect its own data for each species and stock to determine feeding requirements.

Each lot of fish and eggs must be evaluated on its performance from the time it enters to the time it leaves the hatchery. Data should be collected on mortality by phase, food used and purchased, food conversion, and growth rate.

Mortality by Phase. Dead fish must be enumerated and the causes of large losses identified. Significant mortalities must be figured into the feeding equation.

Food Used and Purchased. Food purchased but not utilized is waste. When a surplus or shortage is evident, the hatchery manager must notify the regional hatchery manager. Surplus food at one hatchery may be used at another. If surplus food must be disposed of, be sure to record the amount and add the food cost into the production cost of the fish. Record surplus food and food actually fed as separate items; however, only the latter is used for determining food conversion.

Food Conversion. The kilograms of food required to produce one kilogram of fish flesh is called food conversion. Conversion plays a prominent role in determining feeding levels. Under optimal rearing conditions with nutritionally balanced diets, food (wet weight) to flesh (wet weight) conversions of less than one are possible. This is because fish flesh consists of approximately 80% water and a "dry" diet has only 10 to 15% moisture. The conversion achieved on a "dry" to "dry" basis is not as impressive. This fact should make culturists more sensitive to poor conversion, prompting a more conscientious attitude and awareness toward those factors that can be responsible for it, such as:

1) Poor Feeding Practices:
   Erroneous feeding level on a percent body weight basis.
   Wrong hatchery constant. Wrong inventory.
   Poor feed distribution pattern.
   Frequencies; too few feedings.
   Variable size distribution of the fish.

2) Poor Food Quality:
   Overextended shelf life.
   Nutritionally incomplete diet.
   Nonpalatable.
   Poor milling and pelleting by manufacturer.
   Pulverized by rough handling.

3) Fish in poor health or stressed:
   Disease.
   Overloading.
   Crowding.
   Poor water quality.
   Extraneous stress.

4) Seasonal change:
   Photoperiod.
   Water temperature.

Growth Rate. Accurate data on length and weight increases per temperature unit for each stock, phase, season, and hatchery will help you to gain maximal feeding efficiency.

The growth rate of hatchery fish depends on many factors, e.g., temperature, care, species, stock, diet, feeding level, food quality, health, and sexual maturity. The primary factor affecting growth is water temperature. In addition, the best possible care will result in better growth and conversions. Care, to a large extent, is controlled by the fish culturist. This includes feeding practices, sanitation, loading, handling, disturbances, lighting conditions, and other fish husbandry practices.

Energy Efficiency:

The cost of energy is often a significant portion of the operating expenses of a modern hatchery. Site selection for new hatcheries must emphasize gravity-fed or artesian sources of water. Water should not be pumped or aerated beyond what is required for healthy fish. At an established

hatchery, the hatchery manager must know the available, though usually limited, options for reducing energy requirements. One option, for example, may be to purchase a variable-speed pump to replace a single-speed pump at a hatchery requiring different water flow rates at various times.

Labor Efficiency:

The hatchery manager is responsible for utilizing hatchery personnel to their full potential. He should be a teacher as well as a production foreman. Employees must be challenged on a regular basis. Certain parts of the job are more or less automatic; these are the mundane tasks such as handling food, filling feeders, cleaning tanks and raceways, and collecting eggs. On the other hand, an employee should be urged to perform these tasks more efficiently and to become an expert in his field. This requires on-the-job training, a responsibility of his supervisor. People are the hatchery manager's most important asset.

LIFE HISTORIES OF SALMONIDS

The following generalized life histories will acquaint fish culturists with the basic life cycles of cultured salmonids. Certain details of the life cycle of a particular salmonid stock may differ.

There are five species of Pacific salmon (*Oncorhynchus sp.*) in Alaska: chum, pink, sockeye, chinook, and coho. Except for some sockeye which remain in lakes throughout their lives (commonly known as "kokanee"), all salmon are anadromous, i.e., they begin their lives in fresh water, mature in the ocean, and return to fresh water to spawn.

Adult salmon return to their natal rivers and streams from spring to early winter; the exact timing differs among species and stocks. Migrating adults undergo physiological changes. They stop feeding as they approach fresh water and absorb the oils that they have stored in their body. The digestive system degenerates. All males develop hooked snouts; pink and sockeye salmon males also develop humped backs. All salmon lose their silvery sheen and become darker.

Adults usually enter fresh water before their sex products ripen. When ready to spawn, the female chooses a suitable spawning site and digs a nest or redd in the streambed or lake. The male remains in close attendance, courting the female and fending off competing males. When the redd is completed, the female swims over it and releases some of her eggs. The dominant male moves alongside her and releases milt, fertilizing the eggs. As the female enlarges the redd, upstream gravel washes down and covers the eggs. This sequence of redd-building, courting, and spawning is repeated until the sex products of the female are exhausted. The male may service other females, and several "satellite" males may spawn with one female. All Pacific salmon die after spawning.

The total number of eggs that a female possesses, her fecundity, varies with the species, stock, and size of the individual. The time required for hatching is influenced by temperature, species, stock, and dissolved oxygen in the stream. Newly hatched salmon are called sac fry or alevins. Alevins overwinter in the gravel, nourished by the food stored in their attached yolk sac. After yolk sac absorption or buttoning up, they emerge in the spring as fry.

Eggs, alevins, and fry perish for many reasons. Other spawning salmon dig up the eggs; low stream flows and warm weather reduce the amount of oxygen in the water; floods wash out deposited eggs or cover them with a thick layer of silt; exceptionally low temperatures freeze the eggs or alevins during the winter; birds or other fish eat the fry soon after emergence; and diseases take their toll.

Pink and chum fry generally migrate to sea after emerging, but chum fry may remain in fresh and estuarine water for a month or so.

Chinook, coho and sockeye fry generally remain in a stream or lake for one or two years before going to sea, although some may remain in fresh water

for up to four years. Juvenile salmon migrating from fresh water to the sea are known as smolts.

Different species, and even stocks of the same species, spend varying amounts of time (ranging from 14 months for pink salmon to 4 to 5 years for chum and chinook) in the ocean. During their ocean life, salmon range widely throughout the North Pacific Ocean and the Bering Sea.

Chum Salmon (*Oncorhynchus keta*)

Chum salmon are the most widely distributed of the five Pacific salmon species. In Alaska, chum salmon are most numerous in the Southeastern Panhandle, Cook Inlet, and the large Arctic tributaries of the Bering and Chukchi Seas (Hart 1973; Scott and Crossman 1973).

Chum salmon spawn in gravel riffles in a wide range of stream habitats--from the tidal flats of small streams to springs in the headwaters of large river systems, hundreds of kilometers from the ocean. One of the longest known migrations to a freshwater spawning area is made by Yukon River chum salmon. Many swim more than 2,400 km upstream from the Bering Sea (Merrell 1970)!

At spawning time the chum become dark above, dirty red on the sides, and dusky below. There are greenish bars or mottlings on the sides. In females the red coloration is less pronounced. They bury their eggs in the gravel in the late summer or fall. Like all other Pacific salmon, they die after spawning. The eggs hatch in late fall and early winter, and the alevins remain in the gravel until spring. Fry then emerge and migrate directly downstream. They spend a few months in estuaries or near shore before moving into the open ocean. They grow rapidly in the ocean and after 1 to 4 years return to spawn in the stream where they hatched.

In the stream, chum salmon fry are often found with pink salmon fry. They are distinguishable because pink salmon have no parr marks.

Pink Salmon (*Oncorhynchus gorbuscha*)

Pink salmon occur from Southeastern to the large Arctic tributaries of the Bering and Chukchi Seas in Alaska (Hart 1973; Scott and Crossman 1973). Southeastern Alaska may produce up to one half of the state's commercial catch, and commercially important runs do not occur north of Bristol Bay.

Pink salmon spawn in late summer and early fall. As they ripen, their silvery sheen is replaced by dark greenish sides and a white belly. In Alaska, pink salmon typically spawn in the lower reaches of short coastal streams, although many use the intertidal areas of these streams where the eggs are alternately bathed by fresh and brackish water as the tides ebb and flood. In Prince William Sound, for instance, between 50 and 75 percent of the pink salmon fry are produced in intertidal zones (Bailey 1969).

Spawning usually begins in August or September when stream temperatures are about 10°C. The spawning season and the time of fry emergence are related

to the temperature regimes of the streams. Pink salmon tend to spawn earlier in colder streams and later in warmer ones. The eggs hatch from 3 to 5 months after they are spawned (Bailey 1969).

Alevins spend winter in the gravel and are nourished by the yolk. In April or May, they emerge from the gravel as fry and begin their seaward migration. The fry emerge at night and often reach the estuary before dawn. In the first few days, they form schools near the surface of the water. They move out of the estuary and into the ocean within several weeks. Juvenile pink salmon spend their first summer in coastal waters, and then move into the open ocean in September. After approximately a year in salt water, they return to their home stream to spawn.

Sockeye Salmon (*Oncorhynchus nerka*)

In Alaska, sockeye salmon are found in river and lake systems from Kotzebue Sound through the Southeastern Panhandle (ADF&G 1978b). The Gulf of Alaska and Bristol Bay are the most productive of the Alaskan systems (Hart 1973; Hartman 1971).

Each spring millions of adult sockeye salmon leave their feeding grounds in the North Pacific Ocean to return to the lake and stream spawning areas from which they emerged as tiny fry a few years earlier. Sockeye salmon reach the spawning stage at different ages. Most are 3 to 6 years old at this time. As they ripen, their bodies become bright red and their heads bright green.

In Alaska the spawning season for sockeye salmon extends from late July to early October, depending on the location. Spawning occurs in lake inlet and outlet streams and along the gravel beaches of some lakes down to depths as great as 30 m. In most systems, the amount of spawning in lakes is considerably less than in streams, but in certain lakes, e.g., Karluk and Iliamna, spawning is sometimes extensive. In general, spawning coincides with water temperatures of 4.5 to 10°C. Fish breeding in lakes or in their outlets spawn later than those in the inlet streams because lake waters generally cool more slowly in late summer than do runoff waters in lake tributaries. Fish from early and late runs usually do not spawn in the same parts of the system.

Redds may include fine and coarse gravel and even stones 7 to 10 cm in diameter. The spawning sites are usually selected where there is good waterflow through the gravel for the eggs, which hatch during winter or early spring. The sac fry remain in the gravel for several weeks. Emergence usually occurs during the period from April through June. The fry then move into lakes. They migrate during the night, thus reducing the danger of predation by char, sculpin, trout, and birds. Once in the lakes, fry swim in schools for 1 to 4 years.

When they leave the lake, sockeye smolts are physiologically ready for their life at sea. They migrate rapidly downstream, usually during the darkest hours of the night. Sockeye spend 1 to 4 years maturing in the North Pacific Ocean before returning to spawn.

Certain populations of sockeye become landlocked in lake systems. These "kokanee" have adapted their life cycle to fresh water. Like the anadromous sockeye, they die after spawning.

Chinook Salmon (*Oncorhynchus tshawytscha*)

In Alaska, chinook salmon are common from the Southeastern Panhandle to the Yukon River but range as far north as Kotzebue Sound (ADF&G 1978). Chinook salmon are the largest of the Pacific salmon. They may become sexually mature from their second through seventh year, and, as a result, fish in any spawning run may vary greatly in size. Males mature after one or more years at sea, while females mature after 3 to 5 years at sea. In many spawning runs, males outnumber females in all but the 6 and 7 year age groups. Male chinook salmon that mature after spending less than 1 or 2 years in the ocean are commonly called "jacks." Precocious females are extremely rare.

Alaska's streams normally receive runs of chinook salmon from May through July. Chinook salmon often make extensive freshwater spawning migrations to reach their home streams in some of the larger river systems. Yukon River spawners, bound for the extreme headwaters in Yukon Territory, Canada, will travel more than 3,000 km upriver in 60 days!

As the salmon "ripen" in fresh water, they develop a dusky brown, gray or reddish color. Chinook salmon tend to spawn over coarser gravel and in deeper water than other species of salmon. Each female deposits eggs in several redds, which she excavates in relatively deep, moving water. The eggs usually hatch between November and January depending on time of spawning and water temperatures (Scott and Crossman 1973).

The alevins live in the gravel for a few months until they absorb their yolk. The fry wiggle up through the gravel during spring. In Alaska, most juvenile chinook salmon remain in fresh water until the following spring when they migrate to the ocean, although some remain in fresh water for a second year.

Coho Salmon (*Oncorhynchus kisutch*)

In Alaska, coho salmon are most abundant in Southeastern but range as far north as Kotzebue Sound (ADF&G 1978b). They enter spawning systems from August through November and often ascend several hundred kilometers inland. Adults school in ponds, pools, or lakes for several weeks until ripe. Males develop a bright red body and a dark green back. Females become dusky red upon ripening. In Alaska, coho salmon move into shallow tributaries, which have clean gravel and riffle areas, and spawn from September through January.

The eggs hatch in 3 to 5 months depending on water temperature. Fry emerge from the gravel 2 or 3 months after hatching. Upon emerging, the fry form small schools in shallow areas along the shores of the stream. These schools soon break up, and the fry move into pools or slow water areas where they defend territories. Coho juveniles spend 1 to 3 years in the stream before migrating to sea as smolts in early spring. Smolts remain inshore for a few months and then migrate to the open ocean. Coho salmon normally spend 18 months maturing at sea.

Little is known of the ocean life of coho salmon, although tagging in the Gulf of Alaska has indicated that a large number of Southeast Alaska coho move north along the coastline until reaching the vicinity of Kodiak Island, where they follow the Alaska Gyre in a counter-clockwise direction until they return to the stream of their origin.

Rainbow Trout (*Salmo gairdneri*)

Rainbow trout in Alaska are now abundant not only in their original coastal drainage habitat but also in numerous Southeastern, South-central and Interior lakes where they were introduced (ADF&G 1978b). An anadromous version of this trout is called the steelhead.

During late winter or early spring, when water temperatures increase, adult rainbows usually seek out the shallow gravel riffles of a suitable clearwater stream. Parent fish are usually at least 3 years of age, although males may mature earlier. The female uses her body and tail to excavate a depression in the streambed and then releases eggs which are fertilized simultaneously by an accompanying male. The eggs are covered by the female as she enlarges the nest in an upstream direction. Unlike Pacific salmon, rainbow trout may survive two or three spawning seasons.

Hatching normally takes place 2 to 3 months after spawning, depending on the water temperature. After hatching, rainbow fry depend on the attached yolk sac for sustenance. About 1 to 2 months later the fry emerge from the streambed. Their length at emergence may be from 12 to 25 mm. After emerging, the small trout assemble in groups and seek shelter along the stream margins or protected lakeshores. At the end of their first season of growth, their length may vary from 60 to 250 mm depending on the productivity of the environment.

In lake-dwelling populations, most of the fry move into a lake during their first summer or autumn. However, some fry may spend winter in the spawning stream and enter a lake as fingerlings the next spring. In stream-dwelling populations the young move into riffle areas. They remain there during the summer but tend to move into pools during the autumn and winter. This seasonal shift in habitat is particularly found in streams where rainbow trout and coho salmon occur together.

Anadromous rainbow trout are known as steelhead. After the fry emerge from the gravel, they spend 2 to 3 years in fresh water. The time of seaward migration is more closely related to size than age; 15 to 20 cm is the normal range for smolts. They spend 2 to 3 years maturing at sea before returning to spawn.

Cutthroat Trout (*Salmo clarki clarki*)

The coastal cutthroat trout is found in lakes and streams near the ocean throughout Southeastern Alaska and along the coast to Prince William Sound, its northern- and western-most range (Hart 1973). The anadromous version of this trout is commonly called the sea-run cutthroat.

Cutthroat trout generally mature at 2 to 5 years of age with males usually younger than females at maturity. During late winter or early spring, adults spawn after ascending streams from tide water or lakes. Females dig the redds and after spawning cover the fertilized eggs. Females often spawn with several different males and vice versa. Like rainbow trout, adult cutthroats may survive more than one spawning season.

The eggs hatch about 2 to 3 months after spawning. In the spring, after the alevins absorb their yolk, the fry emerge. They may remain in streams or immediately descend to tidewater or lakes. Sea-run cutthroats, after reaching tidewater, remain in estuaries and ascend streams only to prey on juvenile salmon emigrating from fresh water. Cutthroat trout spend at least 1 year in salt water before maturing.

Dolly Varden Char (*Salvelinus malma*)

Dolly Varden are primarily found in coastal Alaska areas throughout the Southeastern Panhandle, northwestward to the tip of the Aleutian Islands, and in Bristol Bay (ADF&G 1978b). The northernmost part of its range is reportedly Point Barrow. As with rainbow and cutthroat trout, both anadromous and non-migratory populations exist in Alaska.

During the fall, when water temperatures are decreasing, mature Dolly Varden spawn in streams flowing into lakes. In Alaska, these char usually mature after 5 or 6 years of age. As with salmon and trout, females dig the redds and cover the fertilized eggs after spawning. Dolly Varden can spawn twice, but very few live to spawn more than that.

Most eggs hatch in winter, 4 to 5 months after fertilization. After the alevins use up their yolk, the fry emerge during April and May at a size of 20-25 mm. Fry may rear in streams for 3 to 4 years before migrating downstream into lakes or estuaries in the spring. (Scott and Crossman 1973).

Anadromous Dolly Varden migrate back and forth from tidewater to the lake they reared in or to a new lake if they reared in a stream. They migrate to the lakes in the fall and overwinter there. When mature, the anadromous and lake Dolly Varden return to their natal stream.

Arctic Grayling (*Thymallus arcticus*)

Arctic grayling are widespread in all clearwater drainages north of the Brooks Range along the western Arctic slope, throughout all interior Alaska drainages, and as far south as the drainages of Cook Inlet. Westward distribution is not completely known but probably extends no further than Port Moller on the Alaska Peninsula (ADF&G 1978b). Grayling have been introduced to selected lakes throughout Alaska.

In Alaska, grayling become sexually mature at the age of 4 or 5, but some males mature at 2 or as late as 6 years of age. As the ice breaks up in small streams during April or May, adult grayling migrate from ice-covered lakes and large rivers to small gravel or rock-bottomed tributaries. Where there are no suitable small streams, spawning sometimes occurs in muddy-bottomed, vegetated pools below rapids. The males are territorial on the spawning grounds and chase smaller intruding males.

No nest or redd is prepared. During spawning the male curves the extended dorsal fin over the female as if to clasp her to him. There is vigorous vibration and sex products are discharged. The eggs are demersal and adhesive. The female may spawn several times in different areas. After spawning, adults return to the lakes or rivers. The eggs hatch in 2 to 3 weeks. Fry begin to feed several days after hatching and grow rapidly. In the fall, juvenile grayling migrate to large rivers and streams to spend the winter (Scott and Crossman 1973).

Inconnu (*Stenodus leucichthys nelma*)

Inconnu, popularly called sheefish, are most abundant in the Kuskokwim and Yukon River drainages and the Selawik-Kobuk drainages of Kotzebue Sound. They are also present in the smaller rivers of Norton Sound and in the Colville River drainage (ADF&G 1978b).

Inconnu in Alaska are separated into five major populations or groups. The Minto Flats and Upper Yukon River populations are year-round residents in the eastern part of Interior Alaska. The Lower Yukon and Kuskokwim populations overwinter in the delta areas of these two large rivers, while the Selawik-Kobuk population overwinters in the brackish waters of Hotham Inlet and Selawik Lake. These latter groups are termed "estuarine anadromous." Upstream migrations from the wintering grounds begin during the period of ice breakup. The migrations may be to feeding grounds, spawning areas, or both. Migrations may last a few weeks to over four months. Inconnu do not feed in the later stages of their spawning migration but subsist on reserves of body fat.

Inconnu have very stringent requirements for spawning habitat, and only a few rivers in Alaska are suitable. The water must be 1.2 to 2.4 m deep with a fast current over a bottom of differentially sized gravel. Spawning occurs in late September and early October, at water temperatures around 4°C, and only during the afternoon and evening. Inconnu do not dig a redd. The females spawn at the surface of the water with the male swimming underneath fertilizing the eggs. The slightly adhesive fertilized eggs fall to the stream bottom where they lodge in the gravel. Unlike Pacific salmon, inconnu may live to spawn again. A fairly rapid downstream post-spawning migration to the wintering grounds occurs, and the spent fish again begin feeding. Development of the eggs proceeds slowly in the cold water, and up to 6 months may elapse before hatching. The fry travel downstream with the spring floods to the extensive delta areas of the large rivers where they feed.

For an Arctic freshwater fish, the inconnu exhibits a rapid rate of growth. Age studies utilizing scales have shown that fish of each population exhibit distinct growth rates, have a different total life span, and reach sexual maturity at different ages. Age at first spawning varies from 5 to 9 years for males and 7 to 11 years for females. Inconnu appear capable of consecutive spawning, but Russian scientists believe that spawning occurs every 2 or 3 years.

WATER

Fish and water are the primary elements of fish culture. All other elements are embellishments upon these two. A water source must be chosen with as much care as a brood stock, and it must be monitored to ensure that its quantity and quality remain sufficient to produce healthy fish.

Quantity

Water flow rates are measured to the nearest liter/min precision for each incubator and rearing container. Measure flow rates at least once every seven days if conditions are stable, e.g., during normal weather cycles. If conditions are unstable, e.g., during flooding or droughts, measure flow rates daily or more often if needed. (The amount of water needed by eggs and fry is discussed in the "Incubation and Rearing" sections of this manual.)

The usual method for measuring flow rate is the "B.S." (bucket and stop-watch) method. Use a large enough container to catch the water so that it fills in a minimum of 15 seconds. The exact volume of the container must be known; calibrate it to make sure! With large rearing units, use calibrated V-notch weirs or measure the velocity and relate it back to change in volume per unit time. Velocity can be measured by timing a stick moving downstream between two points.

Water flow rates must not be too low or too high. Low flow rates will not deliver adequate oxygen or flush away toxic wastes. High flow rates will roll eggs and cause alevins and rearing fish to expend energy that would otherwise add to their growth.

Quality

Water quality standards for salmon aquaculture are presented in Table 1. Measurement methods, frequency, and precision for the most important water quality characteristics are discussed in the remainder of this section. Remember to take measurements at the same time of day and at the same places so that the results may be compared and correlated.

Dissolved Oxygen:

Record dissolved oxygen (DO) readings at least once a week with a minimal precision of 0.5 mg/liter. At some hatcheries, you may be instructed to measure DO more often. Measure the dissolved oxygen concentration in each incubator or rearing container. Take two readings for each container, one at the inflow and one near the outflow.

Take the inflow measurement at the last accessible spot before the water enters the incubator or rearing container. This may be in the headbox or the incubator manifold. The outflow measurement is taken inside the incubator or container adjacent to the outlet.

DO meters with probes work well, but before each use be certain to either air calibrate the meter or calibrate by conducting a Winkler (iodometric)

Table 1. Alaska Department of Fish and Game water quality standards for salmonid aquaculture.

Water Qualities	Standards
Alkalinity	undetermined
Aluminum	<0.01 mg/liter
Ammonia (un-ionized)	<0.0125 mg/liter
Arsenic	<0.05 mg/liter
Barium	<5.0 mg/liter
Cadmium	<0.0005 mg/liter (100 mg/liter alkalinity)
	<0.005 (≧ 100 mg/liter alkalinity)
Carbon Dioxide	<1.0 mg/liter
Chloride	<4.0 mg/liter
Chlorine	<0.003 mg/liter
Chromium	<0.03 mg/liter
Copper	<0.006 mg/liter (100 mg/liter alkalinity)
	<0.03 mg/liter (≧ 100 mg/liter alkalinity)
Dissolved Oxygen	>7.0 mg/liter
Fluorine	<0.5 mg/liter
Hydrogen Sulfide	<0.003 mg/liter
Iron	<0.1 mg/liter
Lead	<0.02 mg/liter
Magnesium	<15 mg/liter
Manganese	<0.01 mg/liter
Mercury	<0.0002 mg/liter
Nickel	<0.01 mg/liter
Nitrate (NO_3)	<1.0 mg/liter
Nitrate (NO_2^3)	<0.1 mg/liter
Nitrogen (N_2^2)	<110% total gas pressure
	(<103% nitrogen gas)
Petroleum (oil)	<0.001 mg/liter
pH	6.5 - 8.0
Potassium	<5.0 mg/liter
Salinity	<5.0 parts per thousand
Selenium	<0.01 mg/liter
Silver	<0.003 mg/liter (fresh water)
	<0.0003 mg/liter (salt water)
Zinc	<0.005 mg/liter
Sodium	<75.0 mg/liter
Sulfate (SO_4^{-2})	<50.0 mg/liter
Temperature[4]	0° - 15°C
Total Dissolved Solids	<400.0 mg/liter
Total Settleable Solids	<80.0 mg/liter (25 JTU)

Note: Synergistic and antagonistic chemical reactions must be considered when evaluating a water source against these criteria.

titration with appropriate modification when interfering chemicals such as iron are present (American Public Health Association et 1971).

DO content should be ≥ 90% of saturation for the specific water temperature at the inflow. During incubation, outflow readings should be ≥ 7 mg/liter. During rearing, outflow readings should be ≥ 7 mg/liter with an allowable content down to 5 mg/liter for 0.5 hour per 24-hour period. Dissolved oxygen content decreases for a short time after feeding.

Temperature:

Generally, measure water temperatures to the nearest 0.5°C daily. Hatcheries using spring water may require readings only weekly, however, while hatcheries with variable water temperatures throughout the day may require a continous temperature monitor (thermograph). Measure temperatures where the water enters the facility and where it leaves an incubator or rearing container. Each hatchery manager must keep records of water temperatures and seek the optimal temperature range of the species and stocks he cultures.

Other Qualities:

Hydrogen ion concentration, also known as pH, should be recorded every seven days to the nearest 0.1 unit. Sample from the same containers from which you take dissolved oxygen readings. Meters with probes work well.

In special situations, e.g., in a water reuse system, you may be asked to take ammonia (NH_3, NH^+) readings. These should be precise to .01 mg/liter. You may also be asked to take carbon dioxide (CO_2) readings. Be precise to 1.0 mg/liter. Chemical test kits or meters with probes are available for determining ammonia and CO_2 concentrations.

SPAWNING

Fish Transport Permits

Mandated by the Alaska Board of Fisheries under Title 5, Alaska Administrative Code, fish transport permits enable ADF&G management and FRED personnel to review all proposed fish and egg transports. This improves communication within ADF&G and controls the transport of live fish and eggs by public and private agencies. Permits must be approved before an egg take. Permit forms are available at all FRED offices.

Brood-stock Evaluation

Brood stock must have characteristics (e.g., run timing, fry emergence timing) that are compatible with the hatchery environment, FRED goals, and ADF&G harvest management plans. This usually presents no problem at a hatchery where the stock spawns in the stream used as the water supply, since the incubation period is essentially equal for both hatchery and wild fish. However, a problem may occur when the water supplies of the hatchery and indigenous stocks differ, as when a donor stock is transplanted to a hatchery.

In selecting a brood stock, ascertain the run timing of the candidate stocks (after transplantation, a donor stock tends to maintain its own run timing regardless of recipient watershed characteristics) and find out if it presents any problems to hatchery and harvest managers. For example, an early spawning chum stock transplanted into a watershed (used as hatchery water supply) of late spawners will produce very early emergent fry that must be reared until springtime. Where the hatchery water supply differs from that of the indigenous stock, make sure that the hatchery can handle the earlier or later emergent fry and subsequent rearing. Communicate with ADF&G fisheries managers and FRED biologists in the area where juvenile fish are to be released prior to changing stocks.

Bacterial analysis should be performed on potential brood stocks until an adequate disease history is attained. The fish should be screened for bacterial pathogens found in Alaska, such as *Aeromonas salmonicida* (furunculosis), *Renibacterium salmoninarum* (bacterial kidney disease), or *Yersinia ruckeri* (enteric red mouth disease).

Parasite analysis also should be performed until an adequate disease history is attained. Abnormally high levels of metazoan parasites may make the stock unsuitable for hatchery use.

Virus sampling should be performed to detect infectious hematopoietic necrosis (IHN) virus. This should be done annually on sockeye salmon considered for brood stock. Details of disease sampling requirements are available from the fish pathology laboratory.

Adult Capture and Holding

At the typical hatchery, large numbers of adult salmonids are routinely captured and held until ripe. The pros and cons of each method for

capturing and holding adults must be considered during planning. Important considerations are watershed characteristics and stress to the brood stock.

Capture:

Adults can be captured with weirs, seines, gill nets, hook and line, or by electric shocker.

Weir. A weir is an instream barrier to fish passage. Weirs are made of wood slats, iron pipe, steel or aluminum conduit, or wire mesh. Steel or aluminum weirs are lighter and, therefore, more easily moved than wood or iron. Wire weirs may gill small fish and are not recommended.

Aluminum weirs are ideal. They are usually constructed of 3.8 or 6.4 cm aluminum angle frame sections that are usually 2.4 m long. Holes for 1.9 to 3.2 cm diameter aluminum conduit are drilled at desired intervals in the top and bottom horizontal frame sections. Sections are secured to 5.1 cm pipe driven into the stream bottom.

Weirs must be designed to withstand extremes of water flow. Weir pickets of iron, wood, steel or aluminum have approximately 65% void space. The void space for wire weirs is more than 84%. Wire weirs are more difficult to clean and repair during high water than others. Generally, a weir design that has worked successfully in a similar watershed should be used.

Weir placement depends on tides, bottom configuration, composition of the stream, and downstream adult holding area. Weir designs may be straight (perpendicular to flow), diagonal (angled across flow), or V-shaped. The "arms" of the V form two leads into its apex, which points upstream. Bank and bottom erosion can be a problem with straight or diagonal weirs, but V-shaped weirs have fewer erosion problems. Rock-filled gabions help stabilize the stream banks. Sand bags often reduce erosion problems on sand, mud, or fine gravel bottoms. At least two rows of sand bags must be placed in front and in back of the weir to prevent erosion.

Traps may be incorporated into the weir. V-shaped openings in the traps allow fish to enter but make it difficult for them to swim back down-stream. These traps are frequently used for chum and pink salmon, which usually ripen within a few days.

Seine. A seine with an extra-heavy solid core lead line may be used to capture fish downstream from a weir. Seines are also useful in capturing brood stock at the mouths of streams or along lake shores. Fast water, snags, and topography make this method of capture impractical in many areas. Seines work best where fish gather in large pools downstream from weirs or in estuaries.

Gill Net. Gillnetting brood stock is usually not recommended because it is labor intensive and may injure the fish and eggs. It is a useful technique, however, when only small numbers of spawners are needed and when other methods are impractical. Gill nets work best in deep holes and fast water, although snags may be a problem. The net should be no longer than 15 m. Otherwise, large numbers of fish may become caught in the net and die before they can be released. Mesh sizes should be comparable to the mesh used

commercially for the desired species. When gillnetting salmon, mesh should be no less than 11 cm to prevent an incidental catch of trout. Fish are removed from the net by cutting the mesh. Females are removed first, then the males.

Hook and Line. Culturists sometimes use hook and line to capture chinook salmon because of their scarcity and large size. A large, weighted, treble hook is suspended from a limber rod with 45 kg test monofilament line or nylon twine. When snagging, care must be taken not to penetrate the abdominal wall of the fish. Other species may also be captured with hook and line, either by snagging or by sport fishing methods, if other methods are not compatible with water conditions or spawner numbers.

Electric Shocker. Adult salmon may be shocked with an electric shocker, but this must be done carefully. This technique has been used successfully for obtaining chum, pink, chinook, and coho salmon in Alaskan waters.

Holding:

Captured fish must be held in containers to ripen unless spawning is imminent. Holding is usually necessary until the desired number of spawners is collected. Males and females are usually held separately and divided again into green and ripe groups. Pink and chum salmon should be held no longer than 10 days in fresh water; coho, chinook, and sockeye salmon may require a much longer holding period. Avoid holding adult salmon in salt water since adult survival and egg viability may suffer. Hormonal injections are sometimes used to accelerate maturation during holding.

Weir. Weirs may be modified in several ways to hold adults. Double and single weirs are the most commonly used variations.

A double weir consists of two weirs--a straight weir upstream and a straight or V-shaped weir downstream. Adults are held between weirs or in traps between weirs until ripe. Fish enter at the downstream end through a V-shaped "one-way" opening, manually operated gates, or at places where several pickets have been removed. Double weirs are sometimes used for holding sockeye, chinook, and coho salmon adults when the holding period is relatively short, i.e., less than 10 days.

Single weir traps may be used with straight, diagonal, and V-shaped weirs. These traps are generally constructed of aluminum pickets staked in a rectangle with an opening, preferably V-shaped, at the downstream side. The weir extends across the stream from downstream side of the trap. If the trap is used with diagonal or V-shaped weirs, the trap must be located at the part of the weir that is furthest upstream. These traps are commonly used for short-term holding of chum and pink salmon.

Pens. Two basic types of pens are used for holding adults: net and picket pens. Each type is useful for short-term holding of any salmon species.

Net pens are boxes with netting on four sides and the bottom, and may be used in conjunction with weirs. Floating styrofoam collars may be attached to the pens. Pens must be secured tightly and placed in slow moving water. Debris must be removed from these pens frequently.

A small, yet practical, net pen measures 2.4 x 1.2 x 0.9 m and has a 2.5 cm diameter pipe (PVC Schedule 80) frame connected by aluminum "speed- rail" elbows (3-way 2.5 cm connectors). Netting is either 0.6 or 1.3 cm nylon mesh. This pen weighs approximately 7 kg but can be weighted with sand bags. This pen is handy for field operations.

Large floating net pens for holding fish in deep pools or deep areas near stream mouths are sometimes required. Pens measuring up to 3.7 x 3.7 m in each division are constructed of 5.1 x 15.2 cm lumber with 1.3 cm mesh nylon netting. A 30%-full sandbag is enough weight for each bottom corner.

Picket pens are basically weir sections formed in a rectangle or square in a stream. These pens may be used in conjunction with double or single weirs.

Raceways and Ponds. Used for short- and long-term holding of adults at hatcheries, raceways require a directional flow of upwelling water to discourage jumping, which can lead to injuries. Smooth raceway walls prevent abrasions and consequent disease problems; aluminum raceways, trowel finished concrete, or coated concrete raceways are recommended.

Raceways should be designed to allow adults to migrate into them. Weir or fence leads with fishways will do the job. Remember that some species of salmon balk at certain fishway designs. Keeping fish calm increases their survival. The fish may be calmed by disrupting the water surface with sprinklers or covering pens or raceways with black visquine.

Several types of ponds are useful for short- and long-term holding of adults. These are circular, Swedish, and rectangular ponds. Water velocities in all ponds and raceways must be low. As with raceways, water must be introduced below the surface. An upwelling flow is required for retangular ponds and a standard circular rearing flow for circular and Swedish ponds. Rectangular ponds, like raceways, are easy to work with. Crowding and moving adults is rather simple, and rectangular ponds, like raceways, are adaptable for volitional sorting of ripe fish.

Ripeness:

With certain salmon stocks, fish will volitionally move upstream or drop back downstream when ripe or nearly ripe. Raceways and rectangular holding ponds may be equipped to allow this volitional migration of adults to the spawning area. With other raceways, ponds, and holding containers, fish are usually crowded toward an area where ripeness is checked. After crowding, the fish may be anesthetized using MS-222 (tricaine methanesulfonate), CO_2, or 2-phenoxyethanol prior to handling; anesthetization is required for all hatchery brood fish that normally live after spawning, e.g., rainbow trout.

Limit handling during ripeness checks and sorting. Frequent handling may result in disease and low survival. Anesthetization of adults prior to handling reduces stress.

Ripe females may be identified by the following:

(1) Body coloration -- Silvery bright fish are usually not ripe. Fish turn color when ripe; the color differs between species.

(2) Belly distension -- Unripe females have tight body walls and are still rather streamlined in appearance. Upon ripening, the belly wall becomes soft and distended behind the pectoral fins. If held by the caudal peduncle (while supporting the head), the stomach configuration will be that of a teardrop. The abdomen will be flaccid with the eggs feeling distinctly separate. Coho and chinook salmon have thick body walls, and it is sometimes difficult to determine their ripeness with this method.

(3) Egg extrusion -- If the female appears ripe by the preceding method, grasp her by the caudal peduncle. Use cotton or wool gloves to get a good grip on the fish. Cradle the fish against your body by extending your other forearm under her belly. Elevate her tail slightly and apply gentle pressure to her belly with your testing hand, starting below the pelvic fins and moving toward the genital pore. If the pelvic fins begin to fold back against the stomach, the fish will fight and cannot be tested until she relaxes and the fins protrude again.

Egg extrusion is not by itself a determining factor for ripeness. Not all eggs in the ovary ripen at the same time. Pink and chum salmon exhibit a deteriorated ovarian mesentery as spawning approaches. Therefore, unripe females may appear ripe by the extrusion of only a few eggs.

This method should not be used frequently for checking fish as once a few eggs have been extruded, water may enter through the vent and cause eggs to harden. When unsure of the degree of ripeness, you may have to sacrifice one or more individuals of the group by cutting them open to verify ripeness.

Again, handle females carefully because broken and bloody eggs do not fertilize.

Male ripeness can be determined by:

(1) Body coloration and kype -- Coloration is distinctive for males and well as females. Males also develop a hooked snout called a kype.

(2) Sperm emission -- Hold the males in the same manner as females. Slight pressure near the pelvic fins in the region of the genital pore will cause sperm emission in ripe males. Do not apply pressure near the pectoral fins where the vital organs are located. Many males will give one shot of sperm and then no more. Therefore, the fish must be sufficiently ripe to guarantee an adequate amount of sperm.

After ripeness is ascertained, adults are segregated by sex. Furthermore, females are separated into two groups: those that will ripen in 3 days or less, and those that will ripen in 4 days or more. Males are separated into ripe and unripe groups. Each hatchery manager determines the most efficient manner in which to sort and handle the broodstock at his hatchery.

Killing and Bleeding

Salmon broodstock are usually killed by a sharp blow between and slightly behind the eyes. When the number of males is critically low they are not killed because they can produce more milt and be used again. A commercial fisherman's billy or an axe handle makes a fine club. When administering the blow, take care not to hit any part of the body other than the head or broken eggs or bloody sperm may result. Pink salmon females may be killed by breaking the spinal column. Hold the fish in its natural swimming position and block the vent with one hand; with other hand, grab the head with the thumb and forefingers around the gill covers and bend the head back.

Brood fish other than salmon are not killed, but are anesthetized, because they may spawn again in future years. These fish include trout, char, grayling, and inconnu.

Female salmon are bled prior to spawning to ensure that no blood mixes with the eggs. Fish must be bled immediately after killing. This is done by making an incision deep into the caudal peduncle to sever the caudal artery, or by pulling out one set of gills. With the caudal incision method, the cut is usually made from the dorsal side of the peduncle, leaving the ventral side uncut and less likely to bleed into the container. If the ventral side is cut, the bleeding is very rapid but care must be taken to keep blood out of the container. After the caudal is cut, place the fish belly up on a slightly inclined rack to bleed with the head elevated. If the gill removal method is used, the fish is placed head down on the rack. One of the gills is removed to initiate bleeding, which occurs very quickly and thoroughly. With either method, the bleeding period should be 5-10 minutes. Never suspend (hang) females tail down or eggs will be lost.

Do not place killed and bled fish in the sun. If no shade is available, they should be covered with wet burlap. Males should be spawned within 20 minutes of death; females within 30 minutes.

Bleeding of females may be unnecessary if individual hatchery data show that egg fertilization with unbled females is high (≥95% fertilized).

Spawning

Conduct spawning out of the sun and rain. Eggs and sperm must be kept cool and dry. The viability of gametes, i.e., the ability of sperm to impregnate eggs and eggs to become fertilized, declines after gametes are removed from the adults. Gamete viability also declines as air temperature increases. Eggs retain viability for a longer time than sperm at a specific temerature (Withler and Morley 1968; Foerster 1965). When fresh gametes yield ≥95% egg fertilization (the norm), >80% fertilization should occur when one or both gametes are stored at 2 to 3°C for up to 24 hours or at 3 to 9°C for up to 10 hours (Fowler and Banks 1981; Foerster 1965; Plosila and Keller 1974; Poon and Johnson 1970; Withler and Humphreys 1967; Withler and Morley 1968). If gamete storage time will exceed 10 hours, then store the gametes at 2 or 3°C (never 0°C). When possible, store either eggs or sperm but not both; if one of the two gametes is fresh, fertilization should exceed that obtained when both eggs and sperm are stored.

Sperm cells are activated by contact with water and ovarian fluid, but once the vigorous swimming activity is triggered it persists for only 10 to 15 seconds. Unfertilized eggs will begin to take on water and harden if left in contact with water. This will prevent fertilization later because the micropyle will close. Before stripping eggs from a female, waste the first few eggs, since they may contain water, urine, or blood.

Incision Stripping:

Stripping eggs from salmon is accomplished by making an abdominal incision from the genital pore anteriorly around the pelvic fins to the transverse septum. In a two-man spawning team, one person holds the fish by hooking a few fingers under the gill cover, and the other person removes the eggs from the body cavity. If one person is taking eggs, the fish may be hooked under the gill cover, steadying the fish between the legs. The drop distance to the receiving containers should be minimal (30 cm) to prevent damage to the eggs. In ripe pink and chum salmon, the ovarian mesentery deteriorates and eggs should fall readily from the body cavity. In other species, some manipulation may be necessary. If it is, do not squeeze the ovary. Gently shake the anterior portion of the skein. Do not push the eggs down with your fingers as this will break them. Do not attempt to take the last few eggs as this can reduce the fertilization.

Strip eggs into a shallow, clean, dry container, such as a plastic dishpan, aluminum pan, or enamel container (never galvanized metal or copper). Eggs may be stripped into a net or colander to allow excess ovarian fluid, body slime, or broken eggs to pass through. If the eggs do not appear normal, i.e., are tight, hard, "glassy," or if there is an abnormal amount of blood, egg breakage, or fluid from the body cavity, then discard these eggs and take no more from that particular female. The person stripping eggs should check their quality. If eggs are broken, bloody, or if an unusual amount of fluid is found in the body cavity of the female, handling and killing methods should be re-examined. If eggs do not flow freely when the belly is slit and many eggs are still held within the ovarian mesentery, the female is not ripe. Make certain the people checking ripeness are doing it properly. These factors can affect fertilization significantly (Burrows 1949).

If the gametes will be transported separately, put the eggs into containers and place them in coolers; during transport, egg temperature should be 2 to 3°C (never 0°C) if stored for more than 10 hours. Egg containers should never touch ice. When eggs from more than one female are placed into containers, the number of females per container should not generally exceed one for inconnu; four for chinook salmon; eight for chum and coho salmon, char, grayling, and trout; and 10 for pink salmon (special procedures are employed for sockeye).

For some smaller egg takes, where each female is critical, each should be spawned into a separate clean container rather than adding eggs from one female on top of those from another. This will prevent an unnecessary loss of eggs from several females if eggs from one happen to contain blood or water.

To spawn males, support the fish with one arm and hold the caudal peduncle. With the other hand, exert slight pressure on the pelvic fin and genital pore area with a "milking" movement of the fingers. The first shot of milt usually contains urine, so discard it. If there is a shortage of males, they should be kept alive; otherwise they are killed by a blow to the head. Males are not bled.

When fertilization does not occur immediately, the sperm is funneled into whirl-pack bags (maintain a 3:1 air space-to-sperm ratio). For on-site fertilization, the bags should be filled, sealed, and cooled (5° to 9°C) until use.

Live Spawning:

All brood fish that normally live after spawning are anesthetized prior to the egg take. The fish are not killed or cut, and care is taken so that they can spawn again the next year. MS-222 is the anesthetic of choice.

The following instructions pertain to the preparation and use of MS-222. For rapid, safe anesthetization, a concentration of 130-260 mg/liter is usually used for brood stock assuming a temperature between 4 and 15°C. With the proper concentrations, anesthetization may safely exceed 10 minutes, although this is rarely necessary and not recommended. Calculate the amount of MS-222 required (see "Treatment Calculations and Special Treatments" p. 61 in Wood 1974.) Measure the pH, temperature, and dissolved oxygen content (DO) of the water. Mix MS-222 with the water and check the pH. If necessary, buffer the solution with sodium bicarbonate ($NaHCO_3$). The amount of sodium bicarbonate may be one half to two times the amount of MS-222 used and may vary from day to day. Sodium carbonate (Na_2CO_3), which has a wider buffering range, may also be used. Carefully monitor the temperature and DO of the solution. The temperature should never increase more than 5°C above the temperature of the holding or rearing containers and must never exceed 20°C. DO must be $\geq$ 5 mg/liter. Do not contaminate eggs or sperm with MS-222. Useful information on the preparation and use of MS-222 may be found in the ADF&G (FRED) "*Mark/Tag Manual for Salmon* (Moberly et. al. 1977).

After anesthetization, grasp the fish by the caudal peduncle with one hand and support the head with the other hand, keeping the fish level. Dip the fish in clean water to wash off the MS-222.

When hand stripping females, no pressure should be applied anterior of the pelvic fins. The fish should be held belly down in a horizontal position with the head slightly higher than the tail. A gentle upward pressure, applied with the base of the thumb and palm of the hand on one side and fingers on the other, exerted in the region of the pelvic fins will start the flow of eggs. Once started, the flexing of the abdominal muscles of the female is usually sufficient to maintain the flow until most eggs are extruded. To clear the ovaries of eggs, gently apply an upward pressure in the same manner as described previously, continuing posteriorly from the region of the pelvic fin to the genital pore. The process is repeated until

the extremely flaccid condition of the abdomen and the absence of eggs indicates that the ovaries are empty. Care of eggs and sperm, methods of checking for ripeness, and transportation procedures are the same as those described previously.

Another live spawning procedure, air spawning, is used only for female fish and is generally faster and can yield more and cleaner eggs than the hand stripping method (Leitritz and Lewis 1976). This technique is now standard practice on rainbow trout at FRED hatcheries. Females are anesthetized with MS-222. After anesthetization, the female is rinsed in clean water and held in the same way as for hand stripping. A second person inserts a regular point (18 G x 1.5) hypodermic needle (attached to a 478 D Luer-Loc needle adapter) 1.0 to 2.5 cm into the hollow cavity under a pelvic fin. Air pressure not exceeding 210 g/cm^2 (3 lb/in^2) is applied and eggs will fall freely into the container. After spawning, remove the needle and gently purge the air from the fish by hand. Again, care of eggs and sperm, methods of checking for ripeness, and transportation procedures are the same as those described previously.

A minimal "effective" number of 400 spawners is required to maintain genetic variation, which yields a better stock. The male to female ratio is critical to the effective number. This is described in the *Genetics* section of this mannual.

Transporting Gametes

Remote egg takes are common in Alaska. You may be transporting gametes via aircraft, boat, ATV, and backpack.

When shipping gametes separately, place unfertilized eggs into large plastic jars, buckets with lids, aluminum milk cans, or thick-walled plastic bags (double bagged). Ziploc plastic bags are preferred. Never use copper or galvanized containers. Fill the container completely with eggs. This prevents sloshing and possible damage to the eggs. Any container that is kept for reuse must be disinfected. Disposable containers are most efficient.

Place sperm into plastic whirl-pack bags. During large egg takes, the sperm from 10 to 15 males may be placed into each bag. Do not fill the bag over one-quarter of capacity, however, because viable sperm require air. Maintain a 3:1 air-to-sperm ratio.

Place full egg containers and sealed sperm bags in a cooler. Place the bags of sperm so that they lay flat, maximizing the contact of air with sperm. If gamete storage is up to 10 hours, do not exceed 9°C. If storage exceeds 10 hours, then store the gametes at 2 to 3°C (never 0°C). When ice is used, do not allow water from melting ice to touch the egg containers or sperm bags. Transport should not exceed 24 hours or egg fertilization may be less than 80%.

Fertilization

The "dry method" is preferable for both immediate and delayed fertilization. In the dry method, sperm from the desired number of males is combined and

poured onto the eggs. Sperm, eggs, and ovarian fluid are mixed slowly by hand (or feather, if you prefer) or by swirling the sperm and eggs in a shallow container. Ovarian fluid activates the sperm, which are very motile for the first 10 to 15 seconds. Approximately 30 seconds after mixing, add enough water to cover the eggs; mix again slowly. Let the fertilized eggs soak for 30 seconds before rinsing.

Whenever fertilization is delayed, the temperature of the stored gametes must equal the temperature of the incubation water prior to fertilization. This must be done slowly, and may be accomplished by sprinkling containers of gametes with incubator water, or by immersing the containers in water. The water should not touch the gametes themselves. Acclimation usually takes less than 30 minutes (Fowler and Banks 1981; Plosila and Keller 1974).

Rinse fertilized eggs in clean water. Slowly allow water to enter one end of the container and exit the other end. Eggs may be rinsed in baskets either manually or by a flow of water through the eggs. Rinsing is optional. Some hatchery managers have successfully skipped this step by putting fertilized eggs directly into incubators.

During fertilization and subsequent water hardening, water temperatures should not be less then 5°C or more than 12°C.

Water Hardening:

During the water hardening process, the perivitelline fluid, which lies between the yolk membrane (vitelline membrane) and the egg shell (chorion), absorbs water. This causes the egg to increase approximately 20% in volume. The embryo will develop and grow in the perivitelline space, which enlarges at this time. During hardening, the chorion changes from flaccid to turgid. After hardening, the micropyle is closed, and the egg is resistant to further change in shape.

Place the rinsed, fertilized eggs gently into muslin-lined baskets, 19- to 38-liter plastic or stainless steel buckets, milk cans, or incubators for water hardening. Be certain to allow enough space in the containers for egg expansion. Add enough water to the container to allow adequate water absorption by the eggs. If eggs are hardened in flowing water, e.g., in incubators, do not allow the eggs to roll. Do not disturb the eggs while hardening. Most of the water uptake by the eggs occurs within 2 hours, but eggs are often hardened for only 1 hour prior to seeding into incubators or transport. Any movement of fertilized eggs prior to complete hardening must be done very gently.

Transporting Fertilized Eggs:

Water hardened eggs are placed into a cooler on a muslin- or cheesecloth-lined perforated basket. The muslin must be wet and must cover the eggs, but the eggs must not be in direct contact with ice. Ice is needed only on warm days or when transport time exceeds 2 to 3 hours. The temperature within the cooler should not exceed the ambient temperature of the water used for fertilization and hardening.

Eggs should be hardened for 1 to 2 hours prior to transport. Eggs that must be transported before hardening should be placed in 19- to 38-liter plastic buckets or aluminum milk cans filled to the brim with water and sealed shut. The water will cushion the eggs and prevent breakage as long as the container is not excessively jarred. Allow enough space for an egg volume increase of about 20% during hardening; do not fill the container with eggs. Transport should not exceed 12 hours.

Several types of shipping containers are available. Eyed eggs must be moist but not covered with water. Do not ship them in buckets or milk cans; eyed eggs use more oxygen than green eggs and need exposure to air when not in flowing water. Eggs must not be close to hatching when shipped. Transport should not exceed 72 hours.

Sampling Adults

Most data and specimens are collected after the adult fish have been spawned to reduce handling stress and injury. However, count adults at the weir daily. Information obtained from adult fish returning to the hatchery will assist in determining the total number of adults produced by the hatchery.

A spawning crew may be requested to provide otolith or scale samples from the adult spawners for age determination. Information on the percentage of each age class in the spawning population will be used for fecundity calculations. Otolith sampling is time consuming, but it may be necessary when scales have degenerated. Otoliths are located posterior to the eye and dorsal to the anterior part of the gill. Of the three otoliths on each side of the head, only the largest, the sagitta, is taken. To extract it, make an incision laterally along the side of the head and pick it out with forceps. Place the otolith in an envelope or vial for storage and transport.

The Alaska Department of Fish and Game uses two methods for collecting scales. One method utilizes only one scale from each fish and has the advantage of quick collection and reading. Its disadvantage is that, since only one scale is taken, there is a loss of information when individual scales are unreadable.

The second method requires a smear of scales from each fish. The technician responsible for reading the scales can then select two or three good scales from the smear. This method is advantageous in sampling small populations as each piece of information is important. Your selection of a sampling method, therefore, depends on the size of the population. A minimum of 200 samples must be taken to use the easier single scale method. Samples must, of course, be random.

The number of eggs that are available from a female changes as she ripens and may vary by several hundred eggs. Females may lose eggs because of excitement, stream barriers, or redd digging. Ripe females are sampled by the volumetric method of egg counting. This involves taking a sub-sample of a female's eggs, measuring the amount of water it displaces, counting the eggs, measuring the water displaced by the entire ovary content, and

computing the total number of eggs. The fecundity per female and the total number of eggs taken can be computed. Details of the volumetric counting process are given in the *Incubation* section of this mannual.

Length sampling is done with a meter stick. The fish are measured from mid eye to fork of tail to the nearest millimeter. Weigh fish before spawning to the nearest 0.1 kg but avoid using a hanging scale because this could cause egg loss.

Search for fish with clipped or missing fins and enumerate them according to instructions from your supervisor. Fish missing an adipose fin only are probably carrying a coded wire tag. Cut off their heads and send the heads to the mark-tag processing laboratory in Juneau. Be sure to include date and place of capture and mid eye to fork length of the adult with each head. Do not attempt to remove or read the tag in the field unless you were assigned this task specifically.

At remote sites, carcasses may be returned to the stream or lake. At hatcheries, carcasses not sold or given away must be disposed of according to the terms of an Environmental Protection Agency permit.

INCUBATION

Preparation

A hatchery must be thoroughly prepared for incubation. Plans for operations, research, and rearing should be completed before eggs are brought into the facility. The necessary chemicals and equipment should be on hand. Construction that may jar the incubators should cease when the first eggs arrive. Personnel should be briefed thoroughly on the incubation operation and necessary treatments, and each person should know exactly what his tasks are and how they fit into the overall plan.

Employees should be acquainted with the overall stocking plan and any research. Instructions to culturists or technicians should include an explanation of the background for that procedure or research question. This is important both for emergency situations and for the personal education of the culturists.

All incubator loading plans, substrate type, water flow rates, and treatment schedules should be diagramed and displayed. Whether this information originates at the regional office or at the facility, duplicate copies of all planning information should be at both locations. Release, rearing, and marking plans should be completed before the eggs arrive.

Disinfection:

Incubation equipment must be disinfected before each use or when transported to another facility. Remove external dirt and organic material from the surfaces to be disinfected. Immerse equipment in a disinfectant solution made with one of the following:

1. Providone iodine (Betadine) - 200 ppm active ingredient. Percent available iodine is 1.0 for Betadine.

2. Hypochlorite - 200 ppm active ingredient. Household bleach contains 5% active sodium hypochlorite. Active calcium hypochlorite comprises 70 to 74% of the commercial production HTH and 35 to 37% of bleaching powder.

3. Quaternary ammonium (Roccal or Hyamine) - 600 ppm active ingredient. These compounds are quite toxic to coho salmon. Use only if iodine or hypochlorite are unavailable.

See Wood (1974) page 61 for computing the amount of chemical to use. Allow a contact time of at least 30 minutes. Rinse thoroughly.

For large equipment and building sections, the disinfectant solution may be introduced through the spray of a steam cleaner. A solution may be prepared in a 208 liter drum and sprayed with a pump and hose over all surfaces. A pressure-type garden sprayer may be used for smaller equipment.

Incubators

Several types of salmonid incubators are used in Alaskan hatcheries. The five used most frequently are: S40LZ, R29, R30, R48, and Rt16x13H. Incubators are officially designated by three section codes as follows:

Basic Shape	Size (Incubation Dimensions)	Specific Model Code
R = Round S = Square Rt = Rectangle	Nominal surface dimension for containing eggs (in inches)	A one- or two-letter designation, usually the last name initial of the designer or manufacturer.

Incubators used in FRED Hatcheries are:

Official Designation	Common Name
S40LZ	Zenger, NOPAD
R29	302
R30	302
R48	502
Rt16x13H	Heath, Heath Tecna, Heath Tray
R14E	Edo

Receiving Eggs

From the time green eggs are received and until they "eye up," they are very tender. Green eggs are frequently transferred between containers during disinfection and counting before loading them into incubators. When eggs are transferred, they must always be poured under water into receiving containers. Do not pour eggs onto the water surface because some will die.

Fertilization, Water Hardening, and Tempering:

If eggs and sperm arrive separately, fertilization and water hardening must be carried out as soon as the temperature of the gametes is brought close to the hatchery incubation water temperature or when the gamete temperature is raised to at least 5°C. All the water used for fertilization, rinsing, and hardening must be treated with Betadine (100 ppm iodine) before it is expelled through the hatchery effluent to prevent the spread of disease.

If the eggs arrive already fertilized, water hardened, or eyed, the temperature must be checked, and, if necessary, the eggs must be acclimatized to the temperature of the receiving water. This is accomplished by sprinkling the eggs for 30 minutes to 1 hour until the water and egg temperatures are equal. The sprinkling water also must be treated prior to release from the hatchery.

Egg Disinfection:

All eggs brought into a facility from another watershed must be disinfected before exposure to hatchery water. It is recommended that eggs from the

same watershed also be disinfected. Do not disinfect eggs that are within a few days of hatching or premature emergence and dead alevins may result. The Betadine solution should contain 100 ppm available iodine. Buffer the solution with .05% sodium bicarbonate until its pH is the same as the hatchery water. Keep the green or eyed eggs in the buffered disinfectant for 10 minutes. Do not use too thick a mass of eggs or eggs in the center may not receive enough iodine or may not rinse adequately. After exposure, remove the eggs and place them in running water. Raise and lower the baskets to dissipate the disinfectant more rapidly. Allow the water to drain off the eggs for 2 to 3 minutes until clear drops of water fall from the eggs.

Counting Eggs:

An estimate of the number of fertilized eggs must be made prior to loading the incubators. There are several methods for counting eggs. However, at large production hatcheries the number of green eggs placed into incubators for eye-up is roughly approximated (number females spawned x fecundity) with accurate counts made later on eyed eggs. The Burrows' displacement method is often used because of its accuracy, ease, speed, and minimal shock to eggs. The key to this procedure is a determination of the volume of water displaced by the eggs. The eggs, therefore, must be drained adequately to eliminate extraneous water. When the eggs are drained sufficiently, wipe the underside of the basket free of water. Trout and salmon eggs are not harmed by short periods without flowing water.

A perforated dipper is recommended for transferring eggs from the basket to a measuring container. Any residual water clinging to the eggs will be removed in the dipper. You will need a plastic or metal cylinder, 18 cm in diameter and 40 cm in height. It must be equipped with a stopcock and calibrated sight guage and have a capacity of 5 "displacement liters" of eggs. The initial water level is established at the zero reading on the gauge at a depth of approximately 15 cm. The gauge is a standard 25 ml burette that is calibrated in tenths of milliliters.

Pour water into the cylinder and drain the excess through the stopcock until the gauge reads zero. Add eggs until a desired volume is attained. Do not move the cylinder during this procedure. Record the volume of water displaced by the eggs. The measured eggs are poured directly from the cylinder into the incubator.

After this volumetric reading, determine the volume of water displaced by a known number of eggs. Clamp a 50 ml burette to a ring stand and fill it until the water level slightly exceeds 25 ml. Place a sample of 50 eggs on a dry counting trough. The trough is 50 cm long, has a V-shaped bottom, and is curved to a funnel at one end. The counting trough must be dry, because a single drop of water is measureable in the burette and will cause inaccuracies. Just before adding the eggs to the burette, adjust the water level to exactly 25 ml. Add the eggs via the funnel-shaped end of the counting trough, and read the final water level. The final water level must be subtracted from the initial water level to determine the amount of water displaced. For uniformity, all readings in the burette should be made at the bottom of the meniscus.

The accuracy of the transposition between volume and numbers depends upon the selection of random samples. The more numerous the samples, the more reliable the results. One or more random samples should be measured for each volumetric reading made. In small lots, a minimum of five samples should be taken. For large lots of 0.5 million to 2 million eggs, you need not measure a sample for each volumetric reading if precautions are taken to compensate for possible stratification and to ensure randomness.

Do not use the predominant sample in a lot as indicative of the size of the eggs in that lot. This procedure is not statistically sound. Every random sample from a lot of eggs must be included in the calculations to establish an accurate average. The number of eggs per milliliter displaced should be calculated for each sample. The sum of the groups divided by the number of samples will give the average number of eggs per milliliter. The total number of eggs can then be computed by multiplying the average number of eggs per milliliter by the number of milliliters of water displaced by the entire lot (Burrows 1951).

Egg Diameter:

A representative sample of eggs should be measured across the diameter for each run or stock of fish. Standard egg-measuring troughs should be used. Obtain arithmetic means from at least three trough-fulls for each lot sample.

Estimating Percent Fertilization:

It is important to know what percentage of the eggs are fertilized. At each facility, percent fertilization tests should be conducted for each day's egg take. This is extremely vital when new spawning procedures, stocks, or spawning crews are used. Low rates of fertilization may result from faulty egg taking procedures, which must be detected during spawning to allow timely corrections in technique. Experienced culturists expect to fertilize 95% or more of the eggs collected. If a lower rate of fertilization is suspected, percent fertilization tests should be initiated immediately.

Percent fertilization can be estimated by examining a sample of 50 eggs one to two days after fertilization. The early cell divisions in salmon embryogenesis form large cells (blastomeres) that can be distinguished from the germinal disk of unfertilized eggs with low power magnification. To enhance the visibility of embryos, a sample of eggs is soaked in FAA or Carnoy's solution for several minutes. FAA solution is prepared by mixing 30 parts acetic acid, 65 parts of formalin, and 1,000 parts of 50% ethyl alcohol. Carnoy's solution consists of 75 parts of 50% ethyl alcohol and 25 parts acetic acid mixed fresh each time it is used. Soak the eggs in either solution for five minutes. Examine the eggs under a microscope. The unfertilized germinal disk and the embryo of fertilized eggs turn opaque white in the preservative solutions and become visible through the translucent chorion without dissection or staining. A common procedure is to examine the eggs when the four-cell stage is reached. The rate of embryonic development will vary with temperature, species, and possibly with stock.

Loading Green Eggs

Since oxygen consumption and metabolic waste production by eggs are low under normal conditions, space is the limiting factor for loading green eggs. Available space in incubators is commonly designated as both liters of useable volume and liters of green egg volume (displacement liters). Displacement liters of eggs are measured using the Burrows displacement method for egg counting. The displacement value for each type of incubator is the highest volume of eggs that may be eyed successfully in it. A displacement liter of salmon eggs requires approximately 1.45 liters of useable volume; a liter of useable volume is filled by 0.69 displacement liters of eggs (Burrows 1949).

S40LZ:

Before loading the S40LZ incubator with eggs, level the bottom incubator tray. A leveling device is available from the manufacturer. Install an aluminum perforated plate of the correct size for the species into the incubator tray (1 mm diameter holes on 5 mm staggered centers for all Pacific salmon species, except 1 mm diameter holes on 4 mm staggered centers for sockeye salmon). Make sure the plate is well secured with screws so that water will not seep around the edges. Install similar plates in all the trays that will constitute the stack.

Load eggs into the incubator starting with the bottom tray. Eggs can also be loaded after all the trays are stacked by letting them flow from an elevated headbox via tubing into each tray; there is adequate space between trays to do this. Water should be flowing at 57 liters/min through the incubator and the eggs should be loaded under the surface of the water to minimize shock. The maximal number of eggs to load per tray to a depth of 12 cm is approximately 220,000 for chum, king and coho, and 250,000 for pinks. For sockeye, consult the Department's, *Sockeye Culture Policy*.

R29 and R48:

In setting up the R29 and R48 incubators, the aluminum manifold plate (4.6 diameter holes on 76 mm staggered centers) should be bolted in place with the studs provided. Approximately 5 cm of plastic saddles go above the manifold plate.

Secure 2x2 mm mesh fiberglass screen-fabric circle in place on the saddle surface by forming a hoop with a nylon rod and the stainless steel ferrule. Nylon rods will have to be cut to size. The tension of the hoop and ferrule spring will hold the screen in place. The stainless steel tube is designed to hold the nylon rods in a hoop and should accommodate differences in manufacturing tolerances of rod length and incubator diameter. The steel tube, which has a spring to maintain hoop tension, needs a stop at its center so that it will not slide away from the nylon rod joint. This can be accomplished with a pop-rivet through one wall of the tube.

The incubator is now ready for loading. Air bubbles must be kept out of the incubator water. Fill the entire incubator with water and knock out any bubbles before seeding. Water entering the standpipe in a partially filled

incubator will have to fall several inches or feet, thus trapping bubbles under the manifold plate or screen. By initially introducing the water into the egg compartment rather than the standpipe, bubbles may be avoided.

After purging air bubbles, load the eggs gently under the surface of the water. Water flow rates should be 25 liters/min in the R29 (or R30) and 57 liters/min in the R48 incubator. In the R29 (or R30) incubator, the maximal number of eggs to load to a depth of 50 cm is approximately 600,000 for chum, king and coho and 750,000 for pink and sockeye. In the R48 incubator, the maximal number of eggs to load to a depth of 50 cm is approximately 1,700,000 for chum, king and coho, 2,000,000 for pink, and 2,500,000 for sockeye salmon when the incoming water is 90% saturated with oxygen and the temperature is $\leq$ 4.5° C. After loading the eggs, be sure to install the top aluminum pressure plate (same fabrication as the manifold plate).

During incubation, the inlet pipe that passes water from the headbox to the incubator manifold should be just slightly (1-2 cm) under the surface of the water in the manifold. If air bubbles persist, place a plastic tee on the inlet pipe (tee slightly under manifold water surface) to change the vertical plunging force of the water to horizontal force. A net bag of saddles in the manifold will also help trap small air bubbles; make sure that the bag does not plug with debris!

Rt16x13H:

Load the desired number of eggs onto the lower screen in each tray. Cover with the upper screen and fasten the clamp. Use only 2x2 mm mesh size screens. The uppermost tray in the stack is usually left empty as a water filter and degasser.

The maximal number of eggs to load per tray is approximately 10,000 for chum, king and coho, 12,500 for pink, and 16,000 for sockeye salmon when the incoming water is 90% saturated with oxygen and the temperature is $\leq$ 4.5° C. The water flow rate during incubation is 30 liters/min for a 16 tray stack.

Loading Eyed Eggs

Two important limiting factors in loading eyed eggs are space and oxygen consumption by alevins. Space is considered in this section, while oxygen consumption is discussed in the next section on pre-emergent salmon fry carrying capacities. Space is generally the sole limiting factor if the incoming water is 90% saturated with oxygen, the water flow rate is adequate, and water temperature is $\leq$ 4.5°C. The latter is usually the case when fry start emerging at Alaskan hatcheries at natural water temperatures. Oxygen consumption becomes an important limiting factor when the temperature exceeds 4.5°C.

Alaskan hatchery incubation data show that the maximal density for alevins is approximately 675 g/liter of space. Alevins occupy approximately 75% of the available space at this density. The number of eyed eggs to load in commonly used incubators is based on usable space and size of emergent fry as well as the maximal density of alevins. The usable volume of an incubator is simply the area multiplied by the substrate depth. The space

available for alevins is the usable volume minus the substrate volume (saddle substrate is 90% void). Emergent fry sizes (g/fry) are generally 0.39 g for chum, king, and coho fry, 0.26 g for pink, and 0.16 g for sockeye. The highest number of eyed eggs that may be loaded in an incubator is calculated in the equation:

$$\frac{\text{usable volume (liters)} \times \text{substrate void space (0.9)} \times 675 \text{ (g of alevins/liter)}}{\text{mean weight (g) of fry}}$$

For example, the maximal number of chum salmon eyed eggs to load in an S40LZ incubator tray is:

$$\frac{114 \times 0.9 \times 675}{0.39} = 177{,}577$$

The 114 liters of usable volume was determined based on a substrate depth of 11 cm.

Substrate:

Intalox saddles, a plastic rugose substrate, are standard at FRED hatcheries. Emergent salmon fry are larger when they are incubated in a rugose substrate (Leon 1975, 1979 and 1982; Poon 1977). This substrate has proven successful in producing salmon fry that survive well at sea (Kepshire 1982).

The saddles are made of opaque black polypropylene, non-toxic to fish, with a specific gravity of 1.13 ±.03, a 2.38 cm nominal radius, 2.54 cm nominal width, and 4.76 cm nominal length. Each saddle must have four 0.32 cm diameter round holes; each hole must be totally open; i.e., free of any burrs. Saddles must be free of sharp projections and must have rounded corners. Surface area is 207 m^2 per cubic meter. The packing factor is 30-33. Saddles occupy 10% of a given volume, i.e., the void space in a container filled with saddles is 90%.

S40LZ:

To load eyed eggs in the S40LZ incubator, place 10 to 12 cm of substrate on the perforated plate of the lowest tray in the stack and load the desired number of eggs on top. Load succeeding trays in the same manner until the stack is completed. Eggs may also be loaded after all the trays are stacked as for green eggs. Water should be flowing through the incubator during loading, and the eggs should be loaded under the surface of the water. Water flow rates during incubation should range from 57 to 95 liters/min; within this range, reduce the flow rate if air bubble problems occur at high flows. The maximal number of eggs to load per tray is approximately 170,000 for chum, king and coho, 225,000 for pink, and 350,000 for sockeye salmon when the incoming water is 90% saturated with oxygen and the temperature is $\leq$ 4.5° C.

During egg hatching, make sure that the fry weirs are installed or that screens are in the manifolds to catch egg shells and any alevins that might be pushed out of the tray. Decrease the water flow rate during hatching to reduce the latter problem.

R29 and R48:

Complete the procedures described in the first two paragraphs for loading green eggs. For incubators not set up for volitional fry release, do not use center rods or hose clamps. To load eggs, place a layer of saddle substrate on the fiberglass screen followed by a layer of eggs. Then add a second layer of saddles followed by eggs, and then a final third layer of saddles with eggs on top. The maximal depth of substrate is 50 cm when loading the maximal number of 250,000 salmon eggs per R 29 (or R 30) and 725,000 salmon eggs per R 48 when the incoming water is 90% saturated with oxygen and the temperature is $\leq$4.5°C at the initiation of fry emergence. Reduce the substrate depth proportionately if less eggs are seeded. The minimal substrate depth should, however, always be 12 cm. If the number of eggs to load does not exceed 100,000 per R 29 (or R 30) or 250,000 per R 48, then place all the substrate on the fiberglass screen and then load all the eggs on top of the substrate. After loading the eggs, install the top aluminum pressure plate; this plate may be removed during hatching to prevent egg shells from plugging the plate. Water flow rates during incubation should be 38 to 50 liters/min in the R29 (or R30) and 95 to 133 liters/min in the R48.

Rt16x13H:

Load the eyed eggs, cover with the upper screen, and fasten the clamp. The uppermost tray in the stack is usually left empty as a water filter and degasser. See "Loading Green Eggs" for water flow rates and tray capacities.

Pre-emergent Salmon Fry Carrying Capacities

Current models for carrying capacities equate oxygen consumption rate with the hatchery constant feeding rate (Westers and Pratt 1977). Westers (1979) further explains that the feeding rate of all hatchery salmonids is dependent on the temperature-related linear growth rate, the rate of food conversion into fish flesh, and the length of the fish for a given period. This model has been adopted for use in this manual. Emergent fry (pre-feeding fry) receive all of their nutrient requirements from the egg yolk with the exception of dissolved solutes provided in the water. Theoretically, we could then develop a temperature-dependent growth rate (in mm/T.U./day) and a yolk material conversion rate (probably 1.0 or less) for a fry at a given length and water temperature. These values would be applied to an oxygen consumption rate per amount of nutrient yolk material metabolized (g O_2 consumed/kg yolk metabolized).

These empirical values are not presently available, and the hatchery manager will have to collect them for each particular species and incubation method used at his facility. Some known oxygen consumption rates have been applied to a preliminary model that expresses the oxygen consumption rate of emergent fry relative to water temperature.

The model represents a conservative guideline for hatchery use, which becomes very important when water temperatures during fry emergence exceed 4.5°C.

Oxygen consumption rate is represented by a generalized power curve,

$$y = ax^b$$

where y = milligrams of oxygen consumed per kilogram of fish per hour
$a = .00036735$
$b = 3.49$
x = temperature in degrees Fahrenheit which is (°C x 1.8) + 32.

Using the generalized carrying capacity calculation, we can determine conservatively the number of emergent fry that can be held in any incubator as follows:

$$\frac{DO_{in} - 7 \times \text{water flow rate}}{y} = \text{kilograms of emergent fry}$$

$$\frac{\text{kg of emergent fry} \times 1000 \text{ g/kg}}{\text{mean weight (g) of fry}} = \text{number of emergent fry to carry in incubator.}$$

where DO_{in} is the incoming DO concentration in mg/liter; the "7" is the minimal allowable DO content of the incubator's effluent in milligrams per liter.

Water flow rate is liters per hour, i.e., 60 (liters/min),

Y is the milligrams of oxygen consumed per kilogram of fish per hour.

Care of Eggs and Alevins

Eggs may die when handled at certain stages of development. As previously mentioned, eggs are sensitive during water hardening. After that they may be handled until about 12 h after fertilization when cell division begins. They remain tender until just after the eye pigments of the embryos become clearly visible.

Eggs and alevins are very sensitive to other environmental conditions. Excessive exposure to light can kill salmon eggs or cause premature hatching. Alevins exposed to light are abnormally active, which may cause dermal abrasions resulting in fungal infections, white spot disease, yolk sac malformation, and subsequent death due to abnormal organ development, inefficient utilization of the yolk, and premature emergence. Therefore, incubators must be covered with opaque material.

Water Monitoring:

Closely monitor the quality of water entering the incubators during incubation. If water quality falls below the tolerance of the species being incubated, the eggs or alevins will begin dying. Check frequently the water flow rates, dissolved oxygen content, temperature, and pH.

Estimating Developmental Periods:

The rate of development of trout and salmon eggs depends on water temperature, primarily. By monitoring the temperature of the incubation water, one can predict approximately when eggs will "eye up" and hatch and when alevins will "button up" and emerge as fry. Water temperature normally fluctuates throughout the incubation and rearing periods. Therefore, it is necessary to utilize a method of calculating developmental periods that considers average daily temperature. The temperature unit theory developed by Wallich (1900) states that for each species or strain of fish a definite rate of development can be predicted for any temperature between 0° and 15.5°C. Each centigrade degree of the daily average water temperature represents one temperature unit (T.U.). The temperature units are accumulated daily. For example:

Day	Average temperature (°C)	Cumulative temperature units
1	6	6
2	8	14
3	5	19
4	3	22

To plan facility operations, it is necessary to estimate the time sequences for three developmental stages: eyeing, hatching, and emergence. Eyeing occurs when the embryo's eye pigmentation becomes distinct. This coincides with closure of the blastopore when eggs become relatively shock resistant and may be handled for normal hatchery operations. In eggs of Pacific salmon, this stage usually occurs between 250 and 300 T.U.'s. Hatching normally occurs between 450 and 550 T.U.'s for Pacific salmon and at about 350 T.U.'s for rainbow trout (Carlander 1969). At this stage, alevins break out of their egg shell. They remain inactive, lying in the interstices of the substrate while they absorb the yolk sac and continue to grow. Alevins should not be disturbed. Emergence occurs when fry swim to the surface for their initial gulp of air to fill the swim bladder. This usually occurs between 850 and 950 T.U.'s for Pacific salmon and at about 640 T.U.'s for rainbow trout (Carlander 1969).

The number of temperature units required for development is a function of water temperature, species, and stock. Each facility must obtain its own data on development, because the rate will vary. Development will vary according to the severity of the winters, i.e., fry emerge after fewer T.U.'s in cold winters than in warm winters.

Fungus Control:

Aquatic fungi, especially *Saprolegnia parasitica*, are present in most hatchery water supplies and often create obstacles to the successful incubation of fish eggs. Fungi establish themselves on dead organic material. Fungi growing on dead eggs soon smother adjacent live eggs. For this reason, fungus must be controlled through prophylactic treatments with formalin, sodium chloride, or seawater. If you use a commercial product, read the directions, use gloves, and do not inhale formalin fumes. If you

have any doubts about the proper way to handle or use these chemicals, consult your supervisor. Treatments are administered by the drip method. A constant flow siphon (Burrows 1949), I.V. bottle, modified chicken watering device, or a metering pump may be used. Metering pumps are much more accurate and more efficient for larger facilities. Details and calculations for the amount of drug to use are found in Wood (1974).

The frequency of treatment is determined by water temperature, quality, and possible detrimental effects of chemicals on the eggs. Some lots may be protected with only one or two treatments every 7 days; however, in other situations, treatments may be required more often to keep fungus under control. Experience at the hatchery is the best indicator. Subsampling a group of eggs for the presence of *Saprolegnia* is a good way to determine the minimal treatments required to control fungus without disturbing eggs in each incubator. The subsample can be conveniently placed in a small basket hanging inside an incubator adjacent to the water outlet. Frequency of treatments should be kept to a minimum as excessive treatments can cause other problems.

Formalin. Sold as an aqueous solution 37% by weight formaldehyde gas, formalin should be stored in a tightly closed polyethylene or coated metal container at 18 to 21°C. In a constant flow, use 1:500 to 1:1000 (1 to 2 parts per thousand) formalin:water dilution for 15 minutes once or twice every 7 days. Formalin may be toxic to eggs below 1:300 dilution. It has a tendancy to combine with other chemicals and become inactive or more toxic depending on the chemical. Toxicity increases with an increase in temperature. Because of its strong reducing properties, formalin may cause depletion of the oxygen in the water. Make sure that the water is well aerated during treatment. Old formalin may form a white precipitate. This precipitate, paraformaldehyde, is more toxic than formalin and, if prevalent, should not be used. The liquid portion may still be used, but it will be slightly weaker. Formalin is toxic to humans, but a strong odor and irritated eyes usually warn of its presence. It can cause contact dermatitis, so be careful.

Sodium Chloride. Salt (sodium chloride) can be purchased from a scientific supply company or grocery store. Do not use iodized salt; either rock or granulated salt is acceptable. Treat at 9 parts per thousand for one hour.

Following this, stop adding salt and allow the salinity to decrease gradually. Treat once a day or as needed. *Saprolegnia* dies in salt water. Store sodium chloride at room temperature in a tightly closed container. Its shelf life is indefinite.

Seawater. Facilities near seawater can use it for fungus control. Pump the seawater into the headtank for 2 or 3 hours until it replaces the fresh water. At this time, turn off the pump and allow the salinity to decrease. Add the seawater over a few hours to prevent temperature and salinity shock. The temperature of the seawater should be close to that of the fresh water. Treatment should be performed once a day, usually at high tide. Salinity should never exceed 20 to 30 parts per thousand for more than 8 h during each 24-h period. Seawater treatments may need to be supplemented with formalin if high tides do not provide enough seawater.

Shocking:

Dead eggs are an ideal medium for the growth of fungus and must be removed from the incubators to prevent the spread of fungus to live eggs. Physical shock causes dead eggs to turn white. They may then be separated from the live eggs easily.

Shocking causes the yolk membrane of the dead egg to rupture. The proteinaceous globulin inside the yolk precipitates as the salts that held it in solution leak out of the yolk sac. In the live egg, the yolk membrane prevents the diffusion of the salts from the yolk sac to the perivitelline space and from there through the porous shell. Eggs are shocked after eyeing, usually after 300 T.U.'s. At this time the live eggs are hardy enough to withstand the procedure.

Shocking is accomplished by siphoning eggs through 150 cm of hose from the eyeing container to a pail on the floor. The pail should be perforated along the top edge to allow the water to run off without carrying away the eggs. The distance from the end of the hose to the surface of the water in the pail will determine the degree of shock being given. A distance of 20 to 35 cm is generally adequate. The eggs are returned to the incubators following shocking.

Another method is to use a nonimpeller pump to move the eggs directly into an adjacent incubator. In this case the eggs should drop a minimum of 35 cm for proper shocking.

After shocking, the eggs are left for 24 to 48 hours to allow for globulin precipitation. Then they are ready to be picked.

Picking:

Eggs need not be picked unless a 5% or greater mortality has occurred. With adequate fungus control with formulin, a small amount of dead eggs left in the incubator presents no hazard to the live eggs.

FRED Division hatcheries are equipped with automatic egg sorters for picking eggs. These sorters utilize a photoelectric cell to differentiate between opaque and clear eggs. In preparation, siphon the eggs from the incubators into a pail with perforations near the upper rim. Place the eggs into the machine's hopper. After sorting, both the live and dead eggs must be counted. The 1983 model Jensorter egg sorting machine counts live and dead eggs. The electronic fry counter accurately counts live but not dead eggs; this counter may be used in tandem with an egg sorting machine (Kepshire 1983). When automatic counters are unavailable, use the Burrow's displacement method.

Egg Mortality Sampling:

Sometimes it is desirable to know the stage of development at which an egg died. This is done by sampling the dead eggs that are picked off after shocking. Keep dead eggs from each tray or lot separated and labeled to indicate source. Thoroughly mix the eggs in each group before randomly removing a sample. Place each sample in a petri dish with a 10% acetic acid

solution. After the eggs clear (10 to 15 minutes), examine them at 7X magnification on a binocular microscope with transmitted light. Eyed eggs are removed and counted, then each remaining egg is rotated and examined for embryonic tissue.

The eggs are enumerated by category as follows:

1. No development (unfertilized or fertilized).
2. Embryonic development visible but not eyed.
3. Eyed.

Developmental stages of live eggs can be determined by the "percent fertilization" test. Dead eggs and eggs with fungus must be soaked in a 200 ppm chlorine or Betadine solution before disposal, or they must be burned.

Handling Alevins:

Fish culturists should be very gentle with alevins. They are delicate and should not be transferred between incubators except in emergencies. They actually require little attention until the yolk sac is absorbed. However, dead alevins should be removed if possible to prevent the spread of fungus. A small diameter hose may be used to siphon off debris such as egg shells from S40LZ and Rt16x13H incubator trays. Watch the water loss when cleaning incubators.

Fry Emergence

When fry have absorbed the yolk sac or "buttoned up," they swim up from the substrate and begin feeding. Upon emergence the fry must be transferred to rearing containers. In most instances the plumbing allows fry to swim out of the incubator and into rearing containers or holding tanks for counting and evaluation. This is termed volitional migration. In some cases the fry must be moved from the incubators to rearing containers manually. This is called nonvolitional migration, because fry do not leave the incubator on their own. Fry are reluctant to emerge volitionally when the water temperature during alevin incubation is relatively constant (±1°C). Then, the emergence period is too long and results in buttoned up but emaciated fry.

S40LZ incubators are equipped with a fry release gate. At the time of migration, the gate is lifted and fry swim out with the discharge water. At this point the fry along with about 10% of the water are separated out and channeled into the holding tank or rearing container. If fry are transferred manually, each individual tray must be removed from the stack. Fry are loaded into the tank or container. A fry/saddle separator reduces mortality during nonvolitional emergence.

The R29 and R48 incubators may be set up for either volitional migration or nonvolitional transfer. First remove the top pressure plate. For volitional movement, the fry must swim up through the substrate to migrate from the incubator. If fry must be manually transferred from the incubators to holding tanks or fry start tanks, the incubator may be disconnected from the water supply and moved next to or into the holding tank or rearing container.

Rt16x13H incubators are emptied manually by lifting the upper screen in each tray and pouring the fry into containers.

With all other types of incubators except troughs emergence is volitional. Water outlet screens, commonly used to keep pre-emergent sac fry in the incubators, are removed when buttoned up fry show they are ready to migrate.

The date of 50% outmigration is used as the standard to determine the age of fry at emergence. After all fry have emerged from an incubator, the date by which half of them had emerged and the accumulated T.U.'s at that date are recorded.

Fry Counting:

Count volitionally emigrating fry daily. If fry are non-volitionally transferred, count all at once. Methods for counting fry are, in decreasing order of preference, as follows: electronic counting, displacement, and weight counting.

Electronic counting is the most accurate and least stressful to the fry. Northwest Marine Technology, Inc. makes a counter commonly called the Jefferts' counter. The following is a brief description of the unit available at this time. These units are available for use at production hatcheries. Detailed information on operation and maintenance is available from the manufacturer.

The Jefferts' counter consists of an electronics package (waterproof and field serviceable by replacement of a single pull-out board) and counting heads with different size tunnels, which are interchangable to allow use of a single control package for different size fish. Select a tunnel size such that fish must swim through one at a time. The counting speed is theoretically 3.6 million fish per hour, but don't expect it to count more than approximately 120,000 fish per hour. Accuracy of the unit is $\geq$ 98% with proper tunnel size and regulated water flows. Fry tunnel sizes and required water flow rates are as follows:

Diameter of tunnel (mm):	4.8	6.4	7.9
Water flow rate (liter/min):	13.0	23.0	36.0

The correct water flow rate is attained by providing a standard 8-cm head above the counter.

The counter is operated by 12V aircraft or auto battery. The battery needs recharging once a month. The counter is adaptable to a wide variety of layouts and can be inserted into 5 cm I.D. PVC pipe at the outlet of certain incubators. Fry may also proceed downstream from multiple incubators into a trough, which then dewaters the fry for counting at the low flow rates required for small diameter tunnels. The counter easily fits into most hatchery layouts.

The displacement method of counting fish is based on the weight of water displaced by a kilogram of fish. Specific gravity tests show that an average of 1.02 kg of fish displace 1.0 kg of water. Therefore, the total

kilograms of water displaced multiplied by 1.02 equals the kilograms of fish placed in the water. Determine the number of fry per kilogram by sampling for weight. To use this method, mount a sight gauge near the top of a tank. Fill the tank with water as a first step in calibrating the gauge. Water is drawn down to the top of the sight gauge and the level marked on a scale mounted behind the sight gauge. The water is drawn off into a tub and set on a spring or platform balance in the required increments (50, 20, 10, 5 kg, etc.). The gauge scale is marked at the level of each increment. Care must be taken to guard against splashing, and any water accidentally drawn off in excess of the increment should be returned to the tank before marking the scale. The importance of extreme accuracy in this operation cannot be overemphasized as all the fish loaded into tank will be measured on the gauge.

The fish must be dewatered well in order to obtain an accurate fish weight. The error in dewatering small fish will be greater than with large fish, but at this time no exact figures are available. These errors will have to be computed separately for each style of dewatering device and various water flows encountered. When data become available, it will be possible to construct a table with the proper multiplication factors for a specific dewatering device with fish size and water flows as variables.

To use the weight method, first determine the number of fry per kilogram by sampling for weight. A top-loading platform or suspended-type spring balance (scale) should be used for weighing fish. Use a scale precise to the average weight of the fry. In other words, if each fry weighs approximately one gram, the scale should display weights in one gram increments or less. Any support used for suspending a spring balance over a trough or pond should be portable and of a type that leaves the working area entirely clear, with no overhanging projections. Fry can be weighed in a bucket of water that is on or suspended from the scale or directly in a net that is suspended from a spring balance.

After the support and scales are set up, the bucket is hung on the scales. The weighing bucket should be made of a non-toxic substance such as plastic, aluminimum, or steel. Water is added until the desired weight is attained. Fish are then captured in a net, allowed to drain for 5 to 10 seconds and then dumped into the bucket. Passing a hand along the bottom of the net may help to drain the water. The longer the fish drain in the net, the more accurate will be the weight, but anything more than 10 seconds puts undue stress on the fish and should be avoided. What remains is a simple subtraction of the weight of the bucket and water from the weight of the bucket and water plus fish. A scale that can be zeroed or tared is convenient for this type of work. A good written record must be kept throughout the weighing process so that all buckets of fish are accounted for.

Fry Development and Condition:

Fry condition is assessed by calculating the developmental index (K_D) of the unfed emergent fry. The condition factor (K) relates weight to length. Haskell (1959) stated that the body form of fry remains constant for at

least the first 1.5 years. Therefore, the rate of increase is proportional to the increase in length. The general equation describing condition is:

$$K = \frac{\sqrt[3]{W}}{L} \quad \text{or} \quad W = KL^3$$

where W = average weight of the fish in grams,
and L = average fork length in millimeters.

Bams (1970) states that a "condition factor" may be used as a developmental index for unfed fry when modified as follows:

$$K_D = \frac{10\sqrt[3]{W}}{L}$$

where W = weight of fry in milligrams,
and L = fork length in millimeters.

To indicate this special application, the subscript D, for developmental, is added to K. K_D should not be used as an indicator of condition since it is not used with feeding fish.

To calculate the developmental index of fry, take random samples of 25 emergent fry and preserve them in 5% buffered formalin. Unless a specific research project is initiated, these samples should be taken every fifth day after an estimated 10% have emerged and until at least 90% have emerged.

The sample is then placed inside a plastic bag and sealed. Each pack is labeled with the date, incubator number, and observation of the degree of ventral slit closure. The samples are held for a minimum of 6 weeks to allow complete equilibration with the formalin. When measurements are taken, the fry are placed between paper towels to remove excess moisture. No more than five fry should be blotted at one time. This ensures that excessive air drying does not occur. These precautions help to maintain consistency and allow data, which are collected at different times and places, to be compared. Weigh each fry on a Mettler balance to the nearest milligram. The length of each fry is read from the tip of the nose to the fork of the tail. A small plexiglass trough with a millimeter scale glued onto it can be constructed for this purpose. Length should be taken to the nearest millimeter. Handle the fry with forceps only.

Recognizing Disease in Fry:

Diseases affecting fry include infectious hematopoietic necrosis (IHN), bacterial gill disease, white spot (or coagulated yolk), and gas bubble disease.

Viral diseases are characterized by rapid onset and high mortality. Signs of IHN virus disease include exophthalmia (pop-eye), swollen abdomen, lethargy, hemorrhaging at the base of the fins, and a darkening skin color. In sockeye fry, long, opaque, off-white fecal casts trailing from the anus are early signs of IHN.

Bacterial gill disease may involve several species of bacteria. Usually, it is caused by some irritant, such as excess food, which enables bacteria to become established on the gills. Outbreaks may be explosive with high losses within 24 hours, or losses may gradually increase over several days. Signs include fish going off feed, lethargy, pale color, and "riding high" in the water. It can be diagnosed by hatchery personnel. Examine a wet mount of gill arch under a microscope. The presence of long, thin myxobacteria and clubbed gills is indicative of the disease.

Coagulated yolk is characterized by small, white flecks of coagulated yolk in the egg or later in the yolk sac of fry. It is sometimes caused by rough handling of eggs and alevins and excessive treatment with formalin or malachite green. Gas supersaturation can also lead to coagulated yolk disease. The disease may have a genetic component. The incidence of coagulated yolk is more common in salmon incubated without substrate (Emadi 1973).

Gas bubble disease occurs when water is supersaturated with dissolved gases, usually nitrogen and oxygen. Signs of this disease are bubbles under the skin, in fins, tail, mouth and eyeballs, and in the circulatory system. Alevins and recently emerged fry with this disease have bubbles in the yolk, which cause them to float upside down. This disease usually results in death. Gas supersaturation can occur in heated water, well water, spring water, dam water, melted snow water, and pumped water when air enters the pump intake.

REARING

Although releasing young fish as they emerge from the incubators produces acceptable adult returns with some salmon species, notably pink and chum salmon, ocean survivals of all anadromous fishes can be increased by feeding and rearing them at a facility for various periods of time. Pink and chum salmon, which become smolts soon after emergence, are normally reared no more than a few weeks before release. Chinook and coho salmon may be reared for a year or more before release. Sockeye salmon are usually released soon after emergence because of the dangers of infectious hematopoietic necrosis(IHN). Sockeye fry are usually planted in freshwater lakes where they rear naturally and become smolts within two years.

Preparation

Rearing containers and associated equipment must be disinfected before fish are placed in them. Specific chemicals used and concentrations for disinfection are covered in "Incubation." It is not necessary to rinse the chemical off until just prior to use; however, do rinse well. Chlorine at 50 ppm will inactivate virus in 30 minutes when the concentration of organic material is low (0.13%.) Betadine, less affected by organic matter, will inactivate virus in 5 minutes at 50 ppm iodine when organic matter is 1%. A generally safe contact time for effective disinfection is at least 30 minutes for all disinfection chemicals. For higher organic loads, stronger chemical concentrations and longer exposure times are required. Consult the principal pathologist for more details.

For tanks, troughs, raceways, and concrete floors, use brushes or high pressure washers to apply the disinfectant. Combination washer/steamers are also available. For pipelines, hold disinfectant in the lines for 30 minutes.

For complete facility disinfection, all wood in contact with infected water should be burned or replaced. Gravel floors should be flooded with chlorine. For disinfection above water level a formalin fog should be produced. A pesticide fogger is suitable for use with undiluted formalin. Be very careful when using this method and do not breathe the fumes. Conduct these activities with the aid of a senior fish culturist or other experienced culturist.

Earthen ponds cannot be adequately disinfected and should be phased out of operation. Research using hydrated lime and chlorine indicated that neither could eradicate *Myxosoma cerebralis* from earthen ponds. Styrofoam floats cannot be disinfected adequately either. They should be burned when no longer needed or when contaminated.

All rearing containers should be disinfected between lots of fish or at least once a year. Let these containers dry before re-stocking.

Utensils should be disinfected whenever taken to a different pond, raceway, or incubator. Do this by placing them into an iodophor solution after each use and then rinsing them before using again. Minimize movement by having separate utensils for each container if possible.

Rearing Containers

Several types of rearing containers are used at Alaskan hatcheries: troughs, raceways, Swedish ponds, circular tanks, and net pens. Each type has distinct advantages. Troughs and raceways generally do not short circuit water and can provide high water exchange rates at relatively low velocities. Swedish ponds and circular tanks are generally self-cleaning and provide various levels of excercise for the fish. Net pens are inexpensive and require no power output for exchanging water. All rearing containers require your atttention daily.

Troughs:

Rearing troughs are constructed of aluminum or fiber glass. Length x width x height measurements generally range from 2.4 x 0.6 x 0.3 m to 5.5 x 0.4 x 0.2 m. Troughs are used for starting fry and rearing fingerlings. Troughs must be tended daily.

Raceways:

There are several basic types of raceways: land-based horizontal, water-based horizontal, and vertical raceways.

Land-based horizontal raceways have flat bottoms and are constructed of concrete, fiber glass, or aluminum. Some raceways consist of steel frames with plastic liners. Length x width x height measurements generally range from 22.9 x 5.2 x 1.2 m to 36.6 x 3.7 x 1.8 m. Land-based raceways are used for rearing all life stages of salmonids from starting fry to adult brood trout. They should be cleaned at least once every day.

Water-based horizontal raceways are semi-cylindrical floating raceways constructed of nylon-reinforced vinyl (Heard and Martin 1979). Length x width x height measurements of two preferred models are 12.2 x 1.8 x 0.6 m and 18.9 x 4.3 x 2.1 m. These raceways have reared salmon fry and fingerlings to the smolt stage.

Vertical raceways are of two types: floating truncated conical raceways and silos. Conical raceways (Figure 2) are constructed of nylon-reinforced vinyl with small mesh netting at the bottom (Heard and Martin 1979). Water enters the top in a circular pattern and flows out the bottom. A typical conical raceway has a 4-m diameter at the top, 2-m diameter at the bottom and a 3-m depth. Silos (Figure 3) are cylindrical units constructed of fiber glass or steel and are open at the top (Buss et al. 1970). Water enters through a pipe at the center close to the bottom of the cylinder so that the water flows up. Silos range in size (diameter x height) from 208-liter drums (0.6 x 0.9 m) to 20,631-liter units (2.3 x 5.1 m). Both conical and silo raceways are self-cleaning and are used for rearing salmon fry and fingerlings to the smolt stage.

Swedish Ponds:

Constructed of fiber glass, Swedish ponds are square with rounded corners. Water enters along one side at top and exits though a square screen in the bottom center. Water flows in a circular pattern.

Fry and fingerlings can be reared to the smolt stage in these ponds which range in size, i.e., length x height, from 1.1 x 0.6 m to 6.0 x 0.8 m. They are self-cleaning.

Circular Tanks:

Circular rearing tanks are usually constructed of fiber glass or concrete. Circular tanks generally range in size (diameter x height) from 1.2 x 0.8 m to 12.2 x 1.2 m. Water enters along one side at top and exits through a standpipe in the bottom center. Water flows circularly. The tank is self-cleaning when a larger ancillary standpipe with perforations along the bottom surface is placed over the water exit standpipe. Fry and fingerlings can be reared to smolts in these tanks.

Net Pens:

Netting hung on plastic, fiber glass, or metal frames, which in turn are attached to styrofoam or plastic floats, are called net pens. These pens are open on top and may be rectangular, square, circular, or hexagonal. Water flushes through the pens via natural stream or tidal flow. Mesh size of the netting depends on the size of the fish, e.g., chum and pink salmon fry are started in 3-mm mesh while coho fingerlings are reared in a 6-mm mesh. Fry and fingerlings can be reared to smolts in pens.

Small pens may measure (length x depth) 2 x 0.9 m while large pens can exceed 4 x 4 m. Net pens may be cleaned with brushes or an underwater vacuum device. Often fish are transferred to a clean pen so the dirty one can be hauled on land for cleaning.

Removing Solids

Swedish ponds, circular tanks, net pens, and vertical raceways are generally self-cleaning although brushing and vacuuming are necessary at times. Troughs should be cleaned daily by flushing and brushing or by vacuuming.

The rest of this section will pertain only to horizontal raceways. Solids readily settle out in raceways and should be removed at least once every two days by flushing and brushing, flushing alone, or by vacuuming.

Flushing is easily accomplished by rapidly draining the raceway to increase the velocity so that most solids will be pushed out. The fish will also help by stirring up the solids because they have to swim faster. This method will not work well for fry and small fingerlings, which will be forced against the downstream raceway screen by the water.

Without partially draining them, horizontal raceways do not normally have a high enough water velocity to move solids to the downstream end. With flat-bottomed horizontal raceways, baffles are used to increase water velocity at the raceway bottom. Water velocity is determined using the following equation:

$$V = \frac{R\ (L)}{3{,}600}$$

where V is velocity in meters per second,
R is the number of water exchanges per hour,
L is the raceway length in meters, and
3,600 is the number of seconds in one hour.

Most solids settle when the velocity is ± .033 m/s (Jensen 1972). The relationship of velocity to R and raceway length is succinctly presented in Westers and Pratt (1977).

Baffles used in horizontal flat-bottomed raceways will increase the water velocity to move the solids, which makes each raceway essentially self-cleaning (Westers 1979). Baffles are rectangular aluminum plates placed perpendicular to and flush with the sides of the raceway, but not touching the bottom. The top of each baffle is slightly higher than the water depth, therefore all water flows through the gap between the baffle and the bottom of the raceway. The effect of the baffles on the behavior and health of each salmon species is unknown, but baffles apparently do not adversely affect juvenile trout, coho, and chinook salmon. Baffles must be removed when fish are crowded to one end of a raceway as is done when they are moved to other containers or "planted out."

Lateral spacing of baffles and their vertical distance (gap) from the bottom are determined on somewhat of a trial-and-error basis. However, the following recommendations will keep error to a minimum. The velocity under the baffles should be sufficiently strong to carry the solids far enough to be "picked up" by the next baffle. The fish can help this process by keeping the solids stirred up. The larger the fish, the better the job. The suggested velocity under a baffle is from 0.1 m/s to 0.4 m/s depending on the size of the rearing container and fish. To determine this velocity the following equation can be used:

$$V_b = \frac{D_{1(v)}}{D_2}$$

where, V_b is the velocity in m/s immediately behind the baffle,
D_1 is the operating water depth of the rearing container,
D_2 is the gap between the baffle and bottom, and
V^c is the velocity in the rearing unit.

The suggested spacing between baffles is 10 times the velocity under the baffle, e.g., the baffles should be 1 meter apart when the velocity is 0.1 m/s. Generally speaking, the compartments between baffles should be nearly square. Thus a rearing unit with a width of 2 m would have baffles spaced approximately 2 m apart. Again, trial and error must be applied to work out the best arrangement.

Rearing Container Capacities

When rearing salmonids at any life stage, you must ascertain the carrying capacity of each type of rearing container relative to water flow rate and space. The carrying capacity of the water flow, termed "loading," is expressed as kilograms of fish per liter of water per minute [kg/(liters/min)]. The carrying capacity of the container space, termed "density," is expressed as kilograms of fish per cubic meter (kg/m^3).

Loading and density are separate but related characteristics of the rearing container. They relate to each other through the water exchange rate (R) as follows:

$$kg/(liter/min) = kg/m^3\ (.06/R) \text{ or}$$

$$kg/m^3 = kg/(liter/min)\ (R/.06)$$

where R is the number of water volume exchanges per hour in the rearing container. For example, if the volume of the container is 2 m^3 and the water flow rate is 4 m^3/h, then R = 2.

.06 is the volume in cubic meters of a flow of 1 liter/min during a 1 hour period (60 min x 1 liter/min = 60 liters = .06 m^3).

You must know R to calculate loading and density. For example, at R = 1, 1.0 kg/(liter/min) = 16.67 kg/m^3. At R = 4, which is the design exchange rate for new FRED hatcheries (based on Westers and Pratt 1977), a loading of 1.0 kg/(liter/min) results in a density of 67 kg/m^3. Also, at R = 4, a density of 1.0 kg/m^3 results in a loading of .015 kg/(liter/min).

When determining the kilograms of fish to place in a rearing container, calculate the R value first and then the loading to determine the respiratory requirements of the fish. Adequate oxygen must be supplied to the fish before we worry about density.

Exact loadings and densities for safely rearing salmonids are unknown. However, it is very important to know what the approximate maximal loading and density are for each species in each type of rearing container. For example, being too conservative will waste water and space. On the other hand, overloading will stress the fish resulting in poor food conversion, poor growth, and reduced disease resistance and survival (Westers 1979).

The loading and density formulas subsequently discussed in this chapter will be used for all salmon, trout, and char reared in troughs and horizontal raceways. These formulas can work for Swedish ponds and circular tanks under certain circumstances. However, fish are excercised more in circular containers and, therefore, consume more oxygen and excrete more ammonia. Loading is a function of available DO and ammonia in the container water, so circular containers generally have lower loading than a trough or raceway with the same water flow. Do not use these formulas for net pens because the water exchange usually fluctuates drastically during each day. Instead, a safe density for most net pen rearing situations is 8 kg/m^3 for salmon fry and fingerlings.

Loading Equations

Water flowing through a rearing container delivers oxygen and removes metabolic wastes. Oxygen requirements and waste are proportional to the amount of food fed. The flow required for oxygen consumption is determined by knowing how much oxygen the fish require and how much is provided by the water for each liter/min. Though not proven for all salmonid species, a useful general rule is that salmonids require 250 g of oxygen for each

kilogram of food they metabolize (Willoughby et al. 1972). This includes oxygen required for food metabolism, activity, and standard (basal) metabolism. One liter/min of water delivers 1.44 g of oxygen during a 24-hour period for each mg/liter of DO. The available oxygen is the DO level (mg/liter) in the influent less the DO level in the effluent. The rearing container effluent should on the average contain 7 mg/liter DO and must never have less than 5 mg/liter DO; a DO level of 5 to 6 mg/liters is only acceptable for a short time after feeding, i.e., for 0.5 hour per 24-hour period.

Expressed mathematically (Wester 1969), the water flow rate required per kilogram of food fed is:

$$\text{(liters/min)/kg food} = \frac{\text{g required oxygen per kg food fed}}{1.44 \times \text{available oxygen}} \quad \text{or}$$

$$\text{(liters/min)/kg food} = \frac{250}{1.44\ (DO_{in} - DO_{out})},$$

which, expressed as fish loading with the incorporation of the percent body weight to feed daily (% B.W.), becomes

$$\text{(liters/min)/kg fish} = \frac{2.5\ (\%\ B.W.)}{1.44\ (DO_{in} - DO_{out})}$$

The reciprocal of this becomes the loading equation based on oxygen consumption:

$$\text{kg fish/(liters/min)} = \frac{1.44\ (DO\ in - DO\ out)}{2.5\ (\%\ B.W.)}$$

Un-ionized ammonia, which is toxic, is usually the next important loading consideration after dissolved oxygen. Fortunately, most of the ammonia produced by the fish is changed into the harmless (non-toxic) ammonium ion NH_4^+ according to the following reaction:

$$NH_3 + H_2O \rightleftharpoons NH_4OH \rightleftharpoons NH_4 + OH^-$$

The degree of ionization depends primarily on the pH of the water and, to a lesser extent, on the temperature (Emerson et al. 1975) and the salinity (Bower and Bidwell 1978). With each pH unit increase, the un-ionized ammonia level increases approximately ten-fold.

The maximal allowable level of total ammonia in the rearing container effluent produces .0125 mg of un-ionized ammonia per liter (Smith and Piper 1975). The total ammonia production of salmonids is presumably one-tenth of the oxygen consumption (Westers 1979). Therefore, because the oxygen consumption is approximately 250 g/kg food fed, the total ammonia production is approximately 25 g/kg food fed.

The standard loading equation based on ammonia level can be derived the same way as already done for oxygen consumption. The water flow rate required per kilogram of food fed is:

$$(\text{liters/min})/\text{kg food} = \frac{\text{g total ammonia produced per kg food fed}}{1.44 \times \text{allowable level of total ammonia in effluent}}$$

which, because total ammonia in the effluent = .0125 x 100/percent un-ionized ammonia (U.A.) becomes:

$$(\text{liters/min})/\text{kg food} = \frac{25}{1.44\ (1.25/\%\text{U.A.})}$$

where % U.A. equals percent un-ionized ammonia. Simplified, this equation becomes:

$$(\text{liters/min})/\text{kg food} = \frac{25}{1.8/\%\ \text{U.A.}}$$

which, expressed as fish loading with the incorporation of % B.W., becomes:

$$(\text{liters/min})/\text{kg fish} = \frac{2.5\ (\%\ \text{B.W.})}{18/\%\ \text{U.A.}}$$

The reciprocal of this becomes the loading equation based on ammonia production:

$$\text{kg fish}/(\text{liter/min}) = \frac{18/\%\ \text{U.A.}}{2.5\ (\%\ \text{B.W.})}$$

The denominator of this equation is identical to that encountered in the loading equation based on oxygen consumption. Unless pH is high, 18/% U.A. will be greater than 1.44 ($DO_{in} - DO_{out}$). This means that loading based on oxygen consumption is usually greater than loading based on ammonia production. Therefore, accepting the general rule of water aeration to 90% DO saturation, the number of times that the water can be reused between rearing containers is $\frac{18/\%\ \text{U.A.}}{1.44\ (DO_{in} - DO_{out})}$.

Remember again that these equations are valid for troughs, horizontal raceways, and silos. They may be used for conical raceways, Swedish ponds, and circular tanks if:

1) Water velocity does not exceed that in troughs, horizontal raceways, and silos at the same R value, or

2) DO consumption and ammonia production per kilogram of food consumed are measured at the water velocity actually encountered and in the loading equations, substituted for the constants 250 (grams of oxygen required per kilogram of food fed) and 25 (grams of ammonia produced per kilogram of food fed).

Density Equations

After the loading has been determined, the density is then calculated for the available water exchange rate. Using the loading equation based on oxygen consumption, the density is:

$$kg/m^3 = \frac{1.44\ (DO_{in} - DO_{out})\ R}{2.5\ (\%\ B.W.)\ .06}$$

which, simplified, becomes:

$$kg/m^3 = \frac{9.6\ (DO_{in} - DO_{out})\ R}{\%\ B.W.}$$

The maximal densities for each salmonid species that will not adversely affect health are largely unknown. At very high densities, eye and fin erosion problems can be severe, though these problems may be alleviated somewhat with proper rearing container construction and feeding techniques. The density index (DI) concept developed by Piper (1970) is useful because it acknowledges that growth will be reduced if an upper limit of density is exceeded. DI is the kilograms of fish per cubic meter of space per centimeter of body length. In Alaska, DI is 10 for juvenile rainbow trout; 8 for chum, pink, and coho salmon; 6 for chinook and sockeye salmon and 0.6 for rainbow trout brood fish. Keep in mind that lower rearing densities require more rearing space per unit of water flow or loading.

Feeding

Use the "hatchery constant" method of Buterbaugh and Willoughby (1967) to determine feeding levels. The "temperature unit" method of Haskell (1959) has been incorporated into the hatchery constant method to overcome the latter's shortcomings for hatcheries having variable water temperatures. Both methods use fish growth data for each salmonid species at each hatchery to determine feeding levels.

The foundations for using these methods, viz, fish growth, the condition factor, and food conversion, are discussed below.

Growth:

Fish growth is a function of water temperature, with no growth assumed at 0°C (Speece 1973) and at the upper lethal temperature. For trout reared under normal conditions, a definite rate of growth is predictable at temperatures up to 15.5°C according to Haskell (1959). Furthermore, at a constant water temperature, the growth rate in length is constant except when metabolism is altered by factors such as disease or spawning (Haskell 1959). A temperature unit (T.U.) is an average temperature of 1°C over a 24-hour period. As a general rule, the growth rate measured by the growth per T.U. is presumably .05 mm per T.U.

Condition Factor:

The body form or condition factor (K) of a trout presumably remains constant for 1.5 years at a particular temperature and optimal feeding level (Haskell 1959). K relates weight (W) to length (L) in the equation $W = K (L^3)$ which may be rewritten $\sqrt[3]{W} = \sqrt[3]{K}\ (L)$ to show that the cube root of weight is proportional to the length.

K varies with species, stock, food type, feeding level, and other factors, so each hatchery must have its own length-weight tables for calculating condition factors. Until such K values are calculated, use the following values: .008 for chum salmon; .007 for pink salmon; .008 for chinook salmon; .010 for coho salmon; and .011 for rainbow trout.

To determine condition factor, weigh fish in groups of 50 to 100 on a gram scale to obtain total weight. Then measure the fork length of each fish in millimeters, and add them to obtain a total group length. Then calculate the mean weight and length for the group. Fish ≥ 100 mm long may be individually weighed and measured for length to determine individual condition factors.

Feeding levels are based on % B.W., and growth is expressed in terms of length, so we must express the daily percent gain in weight in terms of length. Remember that the cube root of weight is proportional to length, so the daily percent gain in weight is:

$$\frac{\Delta W}{W} \times 100$$

where ΔW is the weight gain and
W is the initial weight.

The daily percent gain in weight expressed as length is:

$$\frac{3 \quad (\Delta L)}{L} \times 100$$

where ΔL is the length gain and
L is the initial length.

The derivation for the latter formula is in Westers (1979).

Food Conversion:

The conversion of food to flesh is an important factor in the feeding equation. It is the ratio of food fed to flesh produced. Thus, if 1.5 kg of food produced 1 kg of fish flesh, the conversion would be 1.5:1. Allowing for factors such as food loss (disintegration in the water), less than optimal water quality, etc., a conversion of 1.5 or less is a reasonable goal for the hatchery personnel.

In theory, conversions of less than 1 are possible due to the high moisture content of fish flesh. The following example for moist diets demonstrates why. First, assume one-third of food goes for growth (Smith 1976), fish flesh is approximately 80% water, and moist diets are approximately 30% water. One kg of food (1,000 g), which consists of 700 g of dry weight of which a third or 233 g goes into growth. The 233 g represents the 20% of the fish flesh which is dry, giving a total of 1,165 g of wet flesh. Therefore, 1 kg of food produced 1.165 kg of flesh, giving a conversion of nearly 0.9.

Temperature Unit Hatchery Constant:

The percent body weight to feed per day (% B.W.) equals the percent gain in weight multiplied by the food conversion. This is calculated in terms of length by the equation:

$$\% \text{ B.W}_L = \frac{3\ (\Delta L)}{L} \times 100 \times \text{conversion} \qquad \text{or,}$$

$$= \frac{300\ (\Delta L)\ (\text{conversion})}{L}$$

Hatcheries with variable temperatures may use growth rate per temperature unit (ΔL/T.U.) multiplied by the average temperature (°C) on that day in place of the daily gain in length (ΔL). The equation becomes:

$$\% \text{ B.W.} = \frac{300\ (\text{T.U.})\ (\Delta L/\text{T.U.})\ (\text{conversion})}{L}$$

Hatcheries with constant water temperatures over periods of time may standardize the numerator of the equation for each period as a hatchery constant (HC), so:

$$\% \text{ B.W.} = 300\ (\text{T.U.})\ (\Delta L/\text{T.U.})\ (\text{conversion})$$

therefore:

$$\% \text{ B.W.} = \frac{HC}{L} = \frac{300\ (\text{T.U.})\ (\Delta L/\text{T.U.})\ (\text{converison})}{L}$$

An average growth rate of .05 mm/T.U. is assumed within the acceptable temperature range for rearing salmonids. This figure should be used at new hatcheries until adequate data are accumulated. It seems that the increase in the growth rate per T.U. (Δ L/T.U.) is a rather gradual one from a low to an optimal temperature, but declines more rapidly as temperatures rise beyond the optimum. Assuming also a conversion of 1.5, the equation becomes:

$$\% \text{ B.W.} = \frac{300\ (\text{T.U.})\ (.05)\ (1.5)}{L} \quad \text{or} \quad \frac{22.5\ (\text{T.U.})}{L}$$

Each hatchery should have its own growth rate data for each species reared. These data will determine more precisely the feeding levels using the "temperature unit hatchery constant" method.

To determine the amount of food to feed the fish, multiply the percent body weight to feed by the weight of the fish, which, as explained previously, can be represented as a function of length.

$$\text{Food to feed} = \frac{300\ (\text{T.U.})\ (\Delta L/\text{T.U.})\ (\text{conversion})\ KL^3}{L} \quad \text{or}$$

$$\text{Food to feed} = \frac{HC\ (KL^3)}{L}$$

The new length (L_n) on each successive day is calculated by adding the L and ΔL values from the preceeding day, i.e.,

$$L_n = L + \Delta L \quad \text{or}$$
$$L_n = L + [T.U. (\Delta L/T.U.)]$$

To determine how much food is needed for each rearing phase, the desired total weight of fish at the beginning of the phase is subtracted from the desired total weight of fish at the end of the rearing phase. This number is multiplied by the conversion rate of food to flesh as follows:

$$\text{Food to feed} = (W_2 - W_1) \times \text{conversion}$$

where W_2 is final desired weight of fish, and

W_1 is initial weight of fish.

The modified hatchery constant method for calculating feeding levels is the best method available today. The feeding levels calculated with this theoretical model should be used as feeding level guides. They must be tempered with common sense and good fish culture practices. Use the feeding period as a time to observe the fishes' effectiveness at eating all of the food presented within a short period. Do not feed more food than will be consumed in five minutes. Be very careful when feeding at extremes in water temperature or fish size. Starting a small fish at low temperatures requires extreme care to avoid over or under feeding. Overfeeding causes gill problems and results in poor conversion. Underfeeding may stress fish and reduce growth and survival. Since our business is producing fish economically, we should care for the fish properly. Proper feeding levels, frequencies and food sizes are important. Spend the time to observe the fish and how they consume the food. Modify the feeding levels to reflect the correct amount to feed. In time you can calculate a hatchery constant that will be accurate for each stock at your facility.

Feeding Frequency:

Feeding frequency is, like feeding level, very important in producing healthy fish and minimizing food waste. The basic rule is the larger the fish, the less frequently per day it is fed. Feed the fish only during the daylight hours, or during a maximum of 12 hours per day in April, May, June, July and August. Recommended feeding frequencies for salmonids reared at temperatures of 5 to 10°C are:

Fish size (g)	Daily feeding frequency
<0.5	30 - 48
0.5-1	15 - 25
1	10
1-?	6
2-5	4-5
5-10	3-4
10-50	2
> 50	1

Production Diet

The Oregon moist diet (OMP II) is the standard production fish food at Alaska Department of Fish and Game hatcheries (a switch to Alaska dry pellet is underway). This diet has proven consistently effective for Pacific salmon and trout at hatcheries throughout the United States for more than 20 years.

Feeding the correct size of food is necessary to prevent starvation or poor food conversion. Recommended OMP II pellet sizes relative to fish sizes are as follows:

Fish size (g)	Food size (diameter in mm)
emergent fry <0.4	starter mash
0.4 - 0.9	0.79 (1/32 inch)
0.9 - 1.8	1.19 (3/64 inch)
1.8 - 3.0	1.59 (1/16 inch)
3.0 - 9.0	2.38 (3/32 inch)
9.0 - 30.0	3.18 (1/8 inch)
> 30.0	4.76 (3/16 inch)
brood stock	6.35 (1/4 inch)

Initial Feeding:

Salmonid fry must be fed as soon as they leave the incubators. During initial feeding, food should be given very frequently but in small amounts. The switch from starter mash to 0.79 mm pellets should occur as soon as possible. Mash irritates gills and reduces survival if the fry are not weaned from it quickly.

Food Storage and Handling:

Fish feeds are perishable products requiring special storage and handling. During storage, several factors may cause destruction or alteration of nutrients.

Oregon moist diet should be delivered at a temperature not exceeding (-)12°C. It must remain in the freezer until just prior to feeding under the following conditions:

1) Maintain the freezer at -23°C,
2) Stagger every other layer to allow air circulation,
3) Keep bags away from the walls, and
4) Place bags on pallets.

Regardless of care, food will degrade in storage. Oregon moist diet has a shelf life of 3 months from the date of manufacture -- not from the time that it is received at the hatchery. Out-of-date food is poor in quality, may be harmful to fish, and, therefore, will not be fed to fish. Careful planning and scheduling will avoid waste and minimize the length of storage.

The most significant loss in food conversion is probably due to the physical condition of the feed, overfeeding or underfeeding. These conditions can be improved. If delivered feed contains too many fine particles, complaints

should be directed to the manufacturer. When unloading, the food should be handled as carefully as possible. Moving bags on a roller conveyor belt is bad practice since this will create a pounding action. Throwing bags around will create more fine particles.

Rearing Container Maintenance

Proper sanitation of rearing units is essential for maintaining healthy fish. Two times each day, inspect screens, baffles, weirs, and drains for damage and accumulation of uneaten food or other debris. Clean these areas often.

Rearing units should be checked for dead fish at least once a day. Since dead fish are often transmitters of disease, they should be removed promptly after discovery. Mortality in each unit should be recorded along with any pertinent observations. Incinerate dead fish, or soak them in a solution of 200 ppm chlorine or iodine (active ingredient) for 12 hours before disposal.

Recognizing Disease

Diseases can be identified before they cause large mortalities by observing the fish for behavioral or physical abnormalities. Feed the fish first thing in the morning and observe them. Look for abnormal swimming, gasping, refusal of feed, listlessness, or resting on the top or bottom of the tank. Regularly, remove a few fish and examine externally for abnormal body color, cloudiness of skin, excess mucus, frayed fins or tail, lesions, protruding eyes, bleeding at the anal vent, or hemmorrhaging. When disease is suspected, try to determine the cause and notify the regional hatchery manager and the fish pathology laboratory.

Sampling

Samples for weight and length measurements are taken routinely on three occasions: at emergence, soon after the receipt of new stock at a facility, and just before release. If practical and applicable, measurements can also be taken once every two weeks for fry weighing less than one gram and once every month for fry weighing more than one gram. Procedures for collecting data must be followed exactly and consistently. The data describe the fish's performance at the hatchery via condition factors, growth rates, and conversions. This information helps establish appropriate feeding levels.

Weight Measurement:

Add water to a container and weigh it. Crowd and mix the fish to obtain a representative sample. Net at least 50 fish, allow them to drain for 5 to 10 seconds, and place them into the pre-weighed container. Record this total weight and subtract the combined container and water weight. Count the fish and then calculate the average weight (g/fish) to three significant figures. Repeat all steps in this paragraph to obtain a total of three samples. The maximal allowable sample weight range is ± .05 x the average weight of the three samples. If the sample range is too large, then take three new samples. A large variation is often caused by inadequate crowding and mixing when sampling. Record all data.

Length Measurement:

Length samples are taken directly after weight samples. Of the fish previously weighed, 50 anesthetized fish are randomly sampled for length measurement. Measure each fish in millimeters (to the nearest millimeter) from the tip of the nose to the fork of the tail. Return the fish to their rearing containers. Calculate the average length per sample and record all data.

Hatchery Effluent

Fish alter the water in which they reside. The effluents that hatcheries discharge consist mainly of water and the products of metabolism. Concentrations of metabolites in the hatchery discharge are proportional to the feeding rate and the water flow rate, and can, therefore, be computed from the following three factors: feeding rate, the food conversion factor (specific for each metabolite), and the water flow rate through the hatchery (Willoughby et al. 1972).

The feeding rate, expressed as kilograms of food per day, is determined from the number of fish, the weight at release, and the percentage of body weight (to feed) by the equation:

$$\frac{\text{kg food}}{\text{day}} > \frac{\text{No. of fish fed (g/fish) (\% B.W./100)}}{1{,}000\ \text{g/kg}}$$

The conversion factor determines the amount of a metabolite produced at any given feeding rate. The conversion factors, derived by Kramer, Chin and Mayo, Inc. (1972), are:

1) NH_3 (Ammonia): .020
2) NO_3 (Nitrate): .020
3) PO_4 (Phosphate): .011
4) Suspended Solids: .370
5) Settleable Solids: .370
6) BOD (Biochemical Oxygen Demand): .420
7) COD (Chemical Oxygen Demand): 1.890

These factors are based on OMP II, except for COD, which is based on dry feed. To determine the amount of a specific metabolite produced daily, multiply the conversion factor by the feeding rate. Conversion factors are given as kilograms of metabolite per kilogram of food. The equation now becomes:

$$\frac{\text{mg metabolite}}{\text{day}} = \left(\frac{\text{kg food}}{\text{day}}\right)\left(\frac{\text{kg metabolite}}{\text{kg food}}\right)\left(\frac{10^6\ \text{mg}}{\text{kg}}\right)$$

Finally, the metabolites produced at any particular feeding rate are related to the hatchery water flow. To determine the flow rate in liters per day:

$$\text{liters/day} = \text{liters/min}\ (60\ \text{min/h})\ (24\ \text{h/day})$$

The final equation is:

$$\frac{\text{mg metabolite}}{\text{liter}} = \frac{\text{mg metabolite/day}}{\text{liters/day}}$$

or combining and simplifying feeding rate, and metabolite production per day equations:

$$\frac{\text{mg metabolite}}{\text{liter}} =$$

$$\frac{10\ (\text{No. of fish fed})(1\ \text{mg/g})(\text{g/fish})(\%\ \text{B.W.})(\text{kg metabolite/kg food})}{\text{liter/min}\ (1{,}440\ \text{min/day})}$$

The water flow rate determines the dilution rate of the hatchery effluent and is the variable that determines the concentration of metabolites in the hatchery effluent.

Because the effluent from rearing containers must have >5 mg/liter DO and .0125 mg/liter un-ionized ammonia, hatchery effluents should be within EPA guidelines for these chemicals. Such is the case in Alaska.

MARKING AND TAGGING

Fish are marked at hatcheries so that biologists can determine their survival rates and contribution to the common property fishery, important information in evaluating a hatchery. Marked fish also provide information on migration routes and can be very helpful to fishery managers.

At FRED hatcheries, fish are marked by clipping off one or more fins. Some are also tagged in the snout with a small coded wire. The wire provides more code combinations than are available from the finclips. In research projects, finclips and coded-wire tags (CWT) are used to evaluate specific techniques and management practices. These projects are generally smaller in scope than production projects.

Planning

The number of fish to be marked should be stated in the facility plans. This number depends on the type of project, how many fish will be released, the expected return, and the recovery effort. Biometricians determine the number of marked fish required.

The Pacific Marine Fisheries Commission (PMFC) keeps records of Pacific North American marks and tags. Representatives from the Alaska Department of Fish and Game and National Marine Fisheries Service coordinate Alaskan marks. Once the need for a mark-tag program is established, a request for marks is sent to the coordinator who allocates clip combinations and codes, and possibly other marks for use at the hatchery. Your technical supervisor can tell you who your mark-tag coordinator is.

Request your finclip assignments before 15 November of the year preceding the fish release. After releasing tagged fish, send relevant information to the coordinator within two weeks. He will record it and forward it to PMFC. The marking and tagging procedures discussed in this section are presented in greater detail in the *Mark-Tag Manual for Salmon* (Moberly et al. 1977)

Marking

Finclipping is a common method for marking fish. Any fin may be clipped except the caudal. All clips handicap the fish and lower its chance for survival. The adipose clip is the least dangerous, however, followed by a single pelvic (ventral) clip. Pelvic fins must be clipped especially close to the base of the fin or it may regenerate. Pectoral clips should be avoided if at all possible. Refer to the *Mark-Tag Manual for Salmon* for finclipping details. The adipose fin is always clipped when a CWT is applied.

Tagging

Coded wire tagging was developed to solve an obvious imbalance, namely, that the number of different finclips available was not enough to satisfy the numerous studies requiring marked fish. All possible single and double clips, excluding caudal clips, yield only 28 different marks. Many of these marks would severely handicap the fish. As you will see later, far more code combinations are available using CWT's.

The tags are injected into the snouts of young salmon by machine. The standard CWT size is 1 mm long and .25 mm in diameter. Half-length tags are also used.

Code Format:

Standard 1 mm tags are marked with three six-digit binary numbers that contain the experimental data. A fourth six-digit binary, called the master, identifies the values of the binary columns, that is, it tells the code reader where the numbers begin. The numbers and master run along the length of the wire. A seventh column, called the column check, provides a check against coding errors in the data. This will be explained later. The coding around a wire tag looks like this:

0	0	1	1	1	1	1
1	1	0	0	0	1	0
1	0	1	1	0	1	1
0	1	1	0	0	0	1

Two things are immediately apparent: the extra digit in the top row, and the use of only two numerals, 0 and 1. The extra half-interval mark in the top row identifies it as the master. It carries no data; its only function is as an identifier. The master number always appears in the pattern shown above, although it may start and end in different places as a result of the randomness of the tag cutting process. So, the master may appear on the tag as 1 1 1 111 0 0, 0 1 1 1 111 0, 111 0 0 1 1 1, 1 111 0 0 1 1, etc. The wire tag is not coded in ones and zeros as we know them. It is coded in notches. A notch is read as "one"; no notch is read as "zero." The notch/no notch columns are spaced 0.12 mm apart.

The notching system is such that tags as short as 0.76 mm will provide unambiguous data. With only two "numerals" to choose from, i.e., "notch" and "no notch," it is necessary to use binary numbers.

Binary Numbers. The number system we commonly use is the decimal system, based on the number ten. The binary system is based on the number two. Because of that, it requires only two numerals (1,0) just as the decimal system requires ten numerals (1,2,3,4,5,6,7,8,9,0).

In decimal numbers, the value of each column is in multiples of 10. For example, 1,350 means 1 thousand, 3 hundreds, 5 tens, and 0 ones. In the binary system, the value of each column is in multiples of 2. The values of binary columns are as follows:

32's	16's	8's	4's	2's	1's
1	0	1	1	0	1

Binary 101101, therefore, is equal to the decimal number 45, that is, 1 thirty-two, 1 eight, 1 four, and 1 one. Counting up to ten in binary is: 1, 10, 11, 100, 101, 110, 111, 1000, 1001, 1010.

CWT's. The numbers on CWT's are notched into the wire by laser. The wire is purchased already coded, and the tag-injector machines merely cut the wire and inject it into the fish. The master number tells the reader in

which direction the numbers are to be read, and identifies the values of the binary columns. This is necessary because the machine often slices through the middle of a number. However, because the numbers are continous along the strand of wire, the entire number can be read. It is somewhat like coming late to a movie and staying to watch the part you missed in the next showing.

The master number and the values of the binary columns it identifies are as follows:

0	0	1	1	1	1	1	MASTER
check	32's	16's	8's	4's	2's	1's	COLUMN IDENTIFICATION

If the number were sliced in half, the master would look like this:

1	1	1	1	0	0	1	1 MASTER
4's	2's	1's	check	32's	16's	8's	COLUMN IDENTIFICATION

The column identified as the check column provides a check against coding errors in the data. It is coded so that the number of notches in any row is always odd. For example, if the data number were 101101, then a correct check bit would be "1." The half-interval identifying notch in the master number is not counted.

Reading the Tag. Do not extract or read tags unless you have been assigned these tasks. To read a CWT, find the master number and orient the tag horizontally so it reads in the correct direction. With the master number properly aligned, rotate the tag so that the master number moves up. The three data numbers will come into view. The order in which they are seen is: Data Number 1, Agency Code, and Data Number 2. The Agency Code identifies the agency, e.g., ADF&G, to which these numbers were issued.

If one were to imagine the surface of the wire unrolled as if it were a sheet of paper, it would look like this (column identification is added in this example for better understanding):

Check	32's	16's	8's	4's	2's		1's	COLUMN IDENTIFICATION
0	0	1	1	1	1	1	1	MASTER NUMBER
1	1	0	1	1	0		1	DATA 1 = 45
1	0	0	1	1	1		1	AGENCY = 15
0	1	1	0	0	1		0	DATA 2 = 50

The information on each of the four "sides" of the tag wire is repeated continuously once every seven spaces. Since tags are cut off every 8.5 spaces, tags may be cut at any point in the number. The same tag cut in the middle would look like this:

4's	2's		1's	Check	32's	16's	8's	COLUMN IDENTIFICATION
1	1	1	1	0	0	1	1	MASTER
1	0		1	1	1	0	1	DATA 1= 45
1	1		1	1	0	0	1	AGENCY= 15
0	1		0	0	1	1	0	DATA 2= 50

Half-Length Tags:

Half-length (0.5 mm) tags were developed for use with small fish (< 1 g). These tags are coded in a four-digit format. The coding scheme is similar to that used in full-length tags. Here is an example:

Check	4's	2's	1's	COLUMN
0	1	1	1	MASTER
1	0	1	1	DATA 1 = 3
1	1	0	1	AGENCY = 5
0	1	0	0	DATA 2 = 4

Note that the master number contains an immediately identifiable half-interval mark. Also, all four rows use the fourth column as a check. The rule is the same as for 1.0 mm tags, i.e., the sum of the notches in each of the rows, including the check bit, is always odd.

Tagging Procedures:

Procedures for tagging are described in the *Mark-Tag Manual for Salmon* (see also Jefferts 1981 and Koerner 1977). When tagging is conducted at remote field stations, the ring magnet assembly from the quality control device may be removed and mounted on a board with a hole in its center. Tagged fish are dropped head first through the magnet into a bucket for magnetization of the wire. Fish are then passed through the field sampling device to check for the presence of a magnetized tag. They must be passed through it upside down with the head exposed and sideways so that the tag is perpendicular to the sides of the detector. Use tally counters to count the fish.

Use two 12-volt car batteries wired in series or a 24-volt aircraft battery to power the tag injector. Remember to plug the positive and negative leads from the batteries into a small adapter box containing a time delay 5-ampere fuse. Run the power cable from the adapter to the tag injector. If the batteries are wired incorrectly, the fuse will blow without damaging the machine. Pack along a supply of fuses.

Checking Tag Retention:

The quality control device (QCD) determines tag loss in fish. It counts the fish, tagged and untagged, which pass through it. Fish lose tags primarily during the first four weeks after tagging. Therefore, tag retention should be determined immediately after the tagging procedure and immediately before release from the facility (rear fish for four weeks after tagging if possible). The field sampling detector (FSD) can also be used for determining tag loss.

If marked and unmarked juvenile fish are held in the same rearing container when checking for tag retention, fish may be netted, anesthetized and checked for adipose clips. Adipose clipped fish are placed in a holding pen inside the rearing container and counted. Normally, 500 adipose clipped fish are sampled for percent CWT loss. This gives a 95% confidence interval of ±2% assuming a 5% CWT loss. CWT loss should not be this great, but a safety allowance is included in this calculation.

The ultimate determination of percent CWT loss results from examination of returning adults. All adipose clipped adults should have CWT's. These fish should be beheaded and the heads sent to the Mark-Tag Processing Laboratory (see "Sampling Adults, page). Do not attempt to remove or read the tag unless you were assigned these tasks.

Instructions for maintaining and troubleshooting all tagging equipment are found in Jefferts (1978 and 1981) and Koerner (1977).

RELEASE

The fish you release represent a year or more of work and worry. The last thing you want is a careless accident on the day of release that may kill what you spent all that time culturing. What follows in this section is a series of release procedures, presented as a guide, to help you avoid such accidents. Release procedures will vary from hatchery to hatchery, but all releases present the potential for error. Aggravating this potential is the brevity of the optimal stocking season, which forces hatchery personnel to transport large numbers of fish in a short time. Haste not only makes waste; in aquaculture, it may make disaster. To use the old analogy, until all your fish are safely released, you are holding a live grenade with the pin pulled. Plan ahead. Think about all the snags that may develop and how you will respond. A difficult release is the acid test for all hatchery managers.

Preparation

A disease inspection is required for all fish before release. Arrangements for this inspection are made with personnel from the pathology section. This imspection must occur at least two weeks prior to release.

Fish are sometimes treated with oxytetracycline (OTC) before release. The OTC is contained in medicated feed. Treatment dose is 10 g of OTC per 100 kg of fish per day for 15 days. If fish are held over for a later release, do not repeat this dose for at least 30 days. The use of certain chemical treatments within 30 days of release may interfere with the normal smolting process. For this reason, always consult your regional hatchery manager and the principal pathologist. No treatments of any kind will be administered within 30 days of release without their consent.

Do not feed your fish for 48 to 72 hours before transit. Fasting reduces the buildup of metabolic waste in the transportation unit, especially if the hatchery water is below 10°C. Remember that the larger the fish, the slower the metabolic rate and the longer the time required to empty the gut.

In hatcheries where water temperatures can be regulated, the hatchery water should be brought as close to the water temperature at the release site as possible. Hatchery rearing temperatures may be significantly higher or lower than the temperature of the receiving water, but an attempt should be made to equalize those temperatures 4 weeks before release.

Release Timing

If young salmonids are released in estuaries, lakes, or streams where there are few natural food organisms, many of them will die. The date of release should be coordinated with the FRED area biologist. Pink salmon fry feed primarily on pelagic zooplankton (Baily 1969; Manzer 1969) and to some extent on epibenthic prey (Kaczynshi et al. 1973). Calanoid copepods are the most important component of their early diets. Harpacticoid copepods are apparently the most important component in the early diet of juvenile chum salmon. Chums are also known to feed on pelagic copepods and larvaeceans during the first few months of ocean life (Bailey et al. 1975; Manzer 1969).

The release of salmonid juveniles should coincide with increasing zooplankton levels to ensure an adequate food supply for the fish. Zooplankton levels can be ascertained by making vertical and horizontal plankton tows in the release area once every 5 to 7 days. An increase in the zooplankton level can be identified by comparison. Depending on the life stage and species of salmonids, the availability of other food organisms, such as insects and shrimp, should be checked.

The temperature of the receiving water is a very important consideration when releasing fish. In much fresh water in Alaska, fish cannot be stocked early in the spring because of ice or in mid-summer because of warm water. Fish generally should not be stocked in waters over 15°C or through an ice cover.

Seawater Challenge Test:

The seawater challenge test is a useful procedure for determining when fish can survive in salt water (Clarke and Blackburn 1977). Each stock (chinook, coho, and steelhead) or release lot of fish must be tested. The seawater challenge test is a relatively new procedure that was developed to measure the osmoregulatory capability of juvenile salmon. If smolting has occurred, fish will be capable of adjusting their plasma sodium level down to the normal value of 170 mM/liter after 24 hours in seawater. If they have not fully smolted, an increase in the plasma sodium concentration will occur and may persist for several days.

Because sodium chloride is the major osmotic component in the extra-cellular fluid, the plasma sodium concentration was chosen to indicate the degree of osmoregulatory competence in juvenile salmon. Another reason for using plasma sodium concentration is the precision and ease with which the sodium ion can be measured. Contact the area and/or regional biologist to set-up this test if you do not already perform it at your hatchery or lack the materials to do so.

Fish exposed to seawater prior to smolting will grow very slowly (if at all) and probably die. Also, smolts held in fresh water too long will revert, i.e., lose their ability to osmoregulate in salt water. Chum and particularly pink salmon fry will generally not attain normal growth if left in fresh water too long. Chum fry may require fresh water for some time after emergence. Pink salmon should be placed into salt water within 10 days of emergence.

Emergent chum fry may require initial fresh water rearing depending on the stock. Before saltwater rearing or release, each chum stock and release lot must be tested for its ability to tolerate salt water. The test will determine how much, if any, freshwater rearing they require. This is very important because it affects rearing costs at hatcheries.

The chum saltwater challenge test is different than the previously described test for smolts. Containers are prepared with salt water at the same temperature and salinity of the seawater rearing site or at 30 parts per thousand salinity. The water temperature must equal or exceed the freshwater incubation and rearing temperature but never exceed 15°C. When the first lot of fry emerge and once every 5 days during freshwater rearing, take 50 to 100 fry and place them into a test container and observe survival and feeding

behavior during the next 5 days. Sample the fry until survival is greater than 95% and the fry are eating as well as those in fresh water. A maximum of five samples will probably be sufficient to determine when the first lot of fry can tolerate salt water.

Pre-Release Counting:

It is important to record the number of fish released from the hatchery. The three best counting methods are, in order of preference, electronic, displacement, and weight counting.

The electronic counter was described in preceeding pages. For salmonids larger than fry, counter tunnel sizes and required water flow rates are as follows:

Diameter of tunnel (mm):	9.5	12.7	19.1	25.4
Water flow rate (liters/min):	52.0	93.0	208.0	371.0

The counter may be set up in a variety of ways. It can be inserted into a screened opening in a raceway, through which fish are crowded, or into a 5 cm I.D. PVC pipe through which fish are passed during transfer from one container to another. It can also be used at the end of a de-watering structure when fish are pumped from one container to another or connected to a single outlet from multiple raceways.

The displacement and weight counting method also have been described in preceding pages.

During volitional release, fish emigrate from the hatchery on their own with little or no interference from hatchery personnel. Volitional release is used when:

1) Fry will not be fed, and the release site is at the hatchery.

2) Smolts are released from raceways at the hatchery when natural food is available and predators are not abundant.

Non-volitional release is the norm for fed fish. Non-volitional release is used when:

1) Fish are transported from the hatchery to other release sites.

2) Fish are released en masse.

Fish are frequently crowded toward the exit of the rearing container for either direct release at the hatchery site or for pumping into transport vehicles. Lowering the water level in the rearing container will help flush the fish out, especially when a crowder is used. Do not overcrowd or crowd the fish too fast or you will stress and injure them. Let the fish move out of the pond at their own pace while slowly crowding them.

Transport

As a prophylactic against disease, all fish transport equipment must be disinfected before and immediately after use in fish transfers or releases.

Nets, buckets, pipes, hoses, boots, and raincoats must be disinfected as well as the transport tank and vehicle. Use a 200 ppm solution of available hypochlorite for 30 min for disinfection (see chemical treatment calculations in Wood 1974).

In tanks with circulating systems, turn the pumps on so the entire system is exposed to the disinfectant. Equipment to be disinfected can be placed directly inside the tank or another container of disinfectant solution for the prescribed time. Thorough flushing of the tank, pumps, and equipment is required before they are used again. Excercise caution in disposing of the disinfectant solution; it may still be toxic to fish. Sodium thiosulfate may be needed to neutralize residual chlorine.

Inspection:

At least two weeks before the expected transport, the necessary equipment must be prepared. The fish pump and transport units should be inspected and serviced. Operate equipment under simulated fish transport conditions. Miscellaneous equipment and supplies, such as oxygen bottles, oxygen regulators, salt, antifoam, pumps, generators, nets, screens, hoses, spare tires, jack and lug wrench, etc., must also be checked.

Filling the Tanks:

The transport tank should be filled with water just before loading with fish. The water should not remain in the tanks for more than 2 hours before loading. Many hatcheries have a special water spigot used only for filling fish transport units. If one is not available at your facility, water should be pumped from the cleanest source available. Do not pump water from the downstream end of your rearing units because this water is probably high in organic matter and low in oxygen.

Chemicals:

The use of salt (NaCl) in transport tanks is proving useful in reducing stress on the fish. A 3 g/liter solution is used at the Ship Creek Hatchery Complex with good success. The addition of salt may reduce stress by making the water more isosmotic with the fish. This isosmotic condition reduces ion loss by the fish, especially phosphate efflux.

The formation of foam and scum can be very detrimental, especially on long trips with a heavy load of fish. Surface foam makes it difficult to observe the fish or the oxygen delivery system during transit. Foam and scum also dry on tank surfaces and must be cleaned off. The formation of foam and scum is reduced with the addition of Dow Corning AF Emulsion. This is a 30% concentration of the Dow Corning Antifoam A with emulsifiers to facilitate mixing it with water. Diluting the emulsion to a 10% solution in warm water

(1 part emusion, 9 parts water) makes it easier to measure and handle. It is best to add the antifoam to the water before adding fish. Add 6.6 ml of the 10% solution for each 100 liters of water. The antifoam normally makes the water slightly turbid.

Oxygen and Circulating Pumps:

Start the oxygen flow to the transport tank at least 10 minutes before the fish are loaded. The first hour of confinement following loading is apparently the critical period of oxygen consumption. An increased oxygen demand, which is caused by stress during loading, occurs during the first 15 minutes of confinement. After the first hour of the trip, the oxygen flow meter setting may be decreased according to the condition of the fish. The oxygen regulator should initially be set for a flow of 10 liters/min and then reduced to 5 liters/min after the first hour of the trip. Personal observation will determine whether the fish need more oxygen.

The circulating pumps or aerators should not be turned on until the fish have been loaded into the tank. This facilitates observation of the fish's condition and is probably less stressful.

A final check is made before leaving to make sure all of the necessary equipment, including an extra oxygen regulator, have been loaded.

Transport Units and Capacities:

Fish size, aeration efficiency, water temperature, fish health, and duration of the trip are all factors which directly affect the weight of fish that can be hauled safely in a fish transport unit. The life support system must meet or exceed the physiological requirements of the fish. The reliability of the system is paramount; if it fails, the fish will quickly die.

When environmental conditions are constant, the carrying capacity of the transport unit is dependant upon fish size. The larger the fish, the lower the relative metabolic rate and the higher the allowable fish density. Capacities are site and stock specific and not merely species specific; therefore, information from other areas can be used only as a guide. Build your own loading capacity tables and graphs when data from your hatchery become available.

Truck Tanks. Truck tanks now used in Alaska vary in size from approximately 1,500 to 15,000 liters (1.5 to 15 m^3). All tanks are equipped with oxygen delivery systems, water circulation systems, aerators, or a combination of these.

To date, maximal capacities successfully used at the Ship Creek Hatchery Complex (1,893-liter fiberglass tanks filled with 1,500 liters of water) are 132 g of chinook or coho smolts per liter (18 g/smolt) in 10°C water for an 8-hour transport, and 150 g of rainbow trout fingerlings per liter (30 g/fingerling) in 10°C water for a 9-hour transport. The tanks used had oxygen and circulating pumps.

A useful guide for determining maximal carrying capacities at different temperatures for king and coho salmon, and rainbow and steelhead trout is found in Westers (1979).

Aircraft. Some waters can be stocked only by aircraft. Special aircraft tanks are manufactured out of fiberglass and aluminum. These tanks are provided with oxygen; however, a pumped circulating system is not provided. The effective capacity of these tanks is probably very near the capacity of regular hauling tanks because of the short time involved in most aircraft hauls. Milk cans are also used to transport fish by air with oxygen supplied through aquarium-type airstones. Helicopters with fire-fighting buckets can also be used for short hauls. Fish can be dropped into a lake or large river pool from altitudes as high as 300 m without injury. Float planes are commonly used for planting fish in lakes or rivers.

Water temperature will rise more rapidly in aircraft tanks, so plan with that in mind. If ice must be used to maintain cool temperatures, make sure the ice is non-chlorinated. Note also that only special green oxygen bottles should be used in aircraft. Watch out for supersaturation with increased altitude on long flights. To date, the maximal capacity used in aircraft containers at the Ship Creek Hatchery Complex is 120 g of coho fingerlings/liter (1.5 g/fingerling) at a final temperature of 15°C for a 7-hour transport.

Small Containers. Fish can be hauled in plastic bags inside insulated boxes or ice chests. Place an empty bag in the box or ice chest. Add water to the bag and pour in the fish. Pump in oxygen and seal the bag with rubber bands. Ice may be added to cool the water during transport. Label the container "Live Fish, Handle With Care" and put a contact person's name and phone number on the container.

Chum salmon fry (0.3 g/fry) have been shipped at 120 g of fry/liter at 4°C with no ice for a 1-hour transport. Grayling and sheefish fry averaging 0.018 g and 0.012 g, respectively, are commonly hauled at 84 g of fry/liter at 4°C for as long as 6 hours.

Loading Fish:

A gravity feed system is the easiest way to load fish but at many of our hatcheries this is impossible. When this method can be used, crowd the fish out of the holding unit into a chute or pipe which passes them over a de-watering device and then into the transport unit.

In most cases, a fish pump with an attached de-watering tower is used for loading transport units. For a facility without a fish pump, the only alternative is to use nets or perforated buckets to move fish from the rearing containers into the transport unit.

System Checks:

As a rule, life support systems should be checked at least once an hour during fish hauling. Oxygen flow should be checked in each tank or compartment, and not just by checking the gauge on the oxygen cylinder. Look into the tanks. Circulating pumps or aerators should also be checked to make sure they are running properly. Alarm systems should be installed on all transport units, but they are not perfect. Make your hourly checks. While checking the life support system, observe the fish. If they gulp at the surface, they are becoming anoxic, and the oxygen flow must be increased.

Release

The temperature of the water in the transport unit should equal that of the receiving water before the fish are released. A 1-hour acclimation is recommended when the temperatures in the transport unit and at the release site differ by more than 10°C.

Do not dump fish into shallow water over rocks or into areas that may be blocked off from the main body of water. Try to park the truck so that water will drain easily; attach a hose to the tank; turn off circulating pumps and aerators; pull stop gate and screen; flush or sweep all fish out of the tank; turn off oxygen; note time. While getting the truck ready to return home, observe the fish to see how they are adapting to their new environment. An estimate of total mortality must be made at this time.

If extremely rough roads will be encountered, add water to the empty transport tank to prevent damage. This will make the unit ride more smoothly.

The transport tank should be rinsed and disinfected as soon as possible. Service the truck as well as the pumps and generators on the transport tank. Change oxygen bottles; make necessary repairs.

If the above procedures are followed, the transport unit will always be ready for the next trip. If, on the other hand, these procedures are not followed, your next transport may be delayed. For example, if the transport unit had to be at a certain place at a specific time in order to plant fish, and you found out while you were filling the unit with water that the oxygen bottles were empty and there was a leak in the unit that would take 2 hours to fix, the trip would have to be cancelled and rescheduled. Remember, no one likes to clean up someone else's mess or fix something someone else has broken. Always keep the transport equipment in good condition.

The fish shipping form is No. 8 of the Standard Production Data Forms. These forms must be submitted after any transport or release of fish or eggs from a FRED facility. People should be familiar with this form before the actual release takes place so that they will know what information must be recorded. The hatchery manager at the facility where the transport originates is responsible for completing and submitting this form. It must be submitted to the office of the regional data coordinator within ten days after release or transport. A copy should also be kept for hatchery records.

FISH HEALTH

Disease Recognition and Action

Whenever abnormal behavior of fish, external abnormalities, or high mortalities occur at a hatchery, an immediate response from the hatchery manager is imperative. Request assistance from your regional hatchery manager and the Fish Pathology Section (FPS) of FRED whenever mortalities appear excessive. An epizootic is occurring when mortalities reach 1.5% per day. This requires immediate attention. A total commitment of the facility and personnel is needed to save the remaining fish. Contact the regional hatchery manager or regional supervisor and FPS immediately.

Mortalities less than 1.5% down to 0.5% indicate that a fish health problem is present. Use routine treatment procedures. Notify the regional hatchery manager and FPS.

Mortalities of less than 0.5% per day should be investigated. Hatchery personnel should attempt to remedy the situation by modifications of environment or feeding. Inform the regional hatchery manager or FPS.

The percentages given above are for total mortalities. It is no less a matter of concern, however, if one lot of fish is dying at 1.5% per day while the others remain healthy. Contact your supervisor immediately and isolate the sick fish as much as possible to prevent transmission of the disease to other lots.

In order to reduce spread of fish disease, make sure that dead fish are incinerated or soaked in a solution of 200 ppm of chlorine or iodine (active ingredient) for 12 hours before disposal.

Preparing Samples

It is often necessary to send fish samples to the pathology lab for analysis. Follow these guidelines when samples are requested. Samples should not be sent except upon request by the FPS. Different procedures are followed for samples for bacteriological, virological, parasitological, and histological analyses. The proper procedures will be provided to hatchery personnel upon initial contact with FPS.

Bacteriology:

Fish must be received either alive or freshly dead on ice. Fish should not be frozen. If transport of fish from a distant site is necessary, then live but dying fish should be bagged in hatchery water with oxygen added. Another sample of live but dying fish should be placed dry in a bag and transported on ice to the laboratory. Shipments should be consigned to a commercial freight delivery company for delivery to the fish pathology laboratory. If the live fish do not survive transport, then the dry fish which will have undergone minimal deterioration and contamination from the water and its bacterial flora will be processed instead.

The number of live fish examined and sampled is normally six; however, this number will vary according to fish size and the problems being experienced at

the hatchery. Samples should be examined from each affected lot, incubator, or rearing container. Consult with FPS for specific sampling requirements in each situation.

Virology:

Methods for collecting samples for virology will vary depending on the type of virus suspected. In general, send sixty moribund fish in a sample. For alevins and fry, send the whole fish; for fingerlings, send the viscera (including kidneys). These must be aseptically removed. For larger fish, aseptically remove the spleen, posterior kidney, liver, and pyloric caecae. Place the samples in sterile bags without water. Do not combine tissues from more than one fish in one bag. Keep samples in the refrigerator or on ice (but not longer than one or two days) or freeze them and then send them to the laboratory as soon as possible.

Parasitology:

For parasitology, fish may be examined shortly after death at the site where they were taken, or they may be preserved for later examination.

Sample six fish showing signs of disease and preserve them in 10% formalin. Specimens more than 10 cm in length should be slit along the belly to ensure formalin penetration. Microscopy for gill parasites should be performed on site.

Histology:

Histological samples should be fixed in Bouin's solution. Fix live fish. Use six moribund fish and six that are apparently normal from the same lot. Dead fish are not suitable for histology. The volume of fixative should be 10 times the volume of the tissue. For fish longer than 10 cm, slit the belly, detach the intestine at the anus and pull the internal organs out slightly. For large fish, send only specified organs in fixative.

Data Collection:

Each sample of fish should be labeled with species, lot, brood year, location, container number, and sample date. In addition, a "Fish/Health Record" is filled out for each lot of fish. Forms are available from the FPS.

Disease Treatment

The regional hatchery manager, the division hatchery pathologist, and the principal pathologist can authorize the use of OMP II (Oregon Moist Pellet) medicated with oxytetracycline. It is used for general prophylaxis, to counter stress of handling or marking, and to counter specific bacterial agents. Sulmet-medicated OMP II can be prescribed by the principal pathologist or the division hatchery pathologist.

Feed can be purchased premedicated, or it can be prepared by adding the appropriate drug to the OMP. Feed is medicated with one of two antibiotics. The first, oxytetracycline (also known as OTC or Terramycin) is recommended for prophylaxis and most bacterial disease treatments. Treat with 10 g/100 kg fish per day for 15 days. The other approved drug is Sulmet

(sulfamethazine), which is fed at 11 g/100 kg fish per day for 15 days. Treatment with medicated food must continue for 15 consecutive days with 30 days between treatments.

When calculating the amount of antibiotic to add to the fish food, it is advisable to reduce the ration fed to the fish. This helps reduce the feeding effort required to build up initial blood levels of antibiotic in the fish. If the fish are still readily accepting feed, the balance of the daily ration may be unmedicated food.

One very good method for applying medication to fish food is a gelatin coat. This delays the leaching action of the water, thus making more medication available to the fish. To accomplish this, dissolve two or three packs of Knox gelatin in 0.95 liters of hot water. Dissolve the proper amount of oxytetracycline in this gelatin mixture. The mixture may be added to the food by surface spraying, mixing in a cement mixer, or by pouring the food back and forth between two tubs until the pellets are evenly coated. Spread the food out on a tarp or sheet of plastic to dry. After it has gelled, feed it to the fish.

Vaccination:

At hatcheries with a history of vibriosis, vaccination may be tried to decrease losses during estuarine rearing periods. Vibrio bacterin is commercially available. Contact FPS for information. Vaccination does not appear to improve adult returns of coho salmon, but it has been very effective on steelhead trout. (Amend et al. 1980.)

Other Treatments:

Regional hatchery managers and hatchery managers can prescribe treatment with seawater or sodium chloride; with formalin for *Saprolegnia* control; with Diquat for bacterial gill disease in coho in freshwater; and, as mentioned before, with medicated feed. All other treatments will be specifically prescribed by the principal pathologist or division hatchery pathologist.

Treatment Methods

Four treatment methods are used commonly: drip, flush, bath, and dip. The drip method is preferred. Treatment levels and dilutions for these four methods are found in Wood (1974.)

Drip Method:

To use the drip method, determine the amount of chemical to be administered (Wood 1974). Determine the liters of water flowing through the fish or egg container each minute. The chemical, in an aqueous solution, is dispensed in a constant flow or drip over the required treatment time. For circulating type containers (e.g., Swedish ponds, circular tanks), first add enough chemical to obtain the desired concentration in the container and then begin the drip treatment. If possible, flush the container at the end of the treatment by dropping the water to the lowest reasonable level and then allow to refill.

Flush Method:

The flush method should be used only in containers that exchange their entire volume of water at least once an hour. Determine the amount of chemical to be administered (Wood 1974) and add the chemical at the intake end of the container. This method is not recommended at FRED hatcheries.

Bath Method:

Again, determine the amount of chemical to be administered (Wood 1974). Lower the water level in each container to 15 cm in depth for every 180 to 220 kg of fish. Halt the flow of water. Add and thoroughly mix the chemical. Use standby water pumps for aeration and circulation of the water. After the prescribed treatment time, start water flowing in the container again and rapidly flush out the chemical. Repeat treatments daily if necessary. This method is not recommended at FRED hatcheries.

Dip Method:

Add and thoroughly mix the prescribed amount of chemical (Wood 1974) to a 45-liter container of water. Containers of plastic, fiberglass, stainless steel or aluminum are recommended. Immerse a net full of fish into the container for 5 to 10 seconds. Change the solution after 8 to 10 nets of fish. This method is not recommended at FRED hatcheries.

Be certain to flush out the chemicals as quickly as possible at the end of the treatment time when using the drip, flush, or bath methods.

Chemicals must always be handled with care. Always read the label directions, follow all precautions, and know the antidotes. Potentially dangerous chemicals should always be removed from the hatchery effluent prior to discharge into watersheds.

GENETICS

Operating a hatchery contrary to sound genetics procedures is like driving a car and never refueling it. Sooner or later both the car and the brood stock will run out of gas. The long-term effects of poor genetics procedures at a hatchery are not as immediately apparent as a disease epizootic, but they can be just as devastating.

To continue the automobile analogy, the brood stock's "gas" is its genetic diversity. Genetic variation is extremely important to a population. It is required for adaptation to a changing environment and is important in such traits as disease resistance. Geneticists have found that a reduction in genetic diversity reduces the ability of a stock to breed and reproduce. Genetic diversity can be maintained by following the guidelines below for brood stock sample sizes.

FRED's geneticist develops genetic profiles of salmonid stocks that are used to gauge the amount of diversity in and between stocks. The profiles are developed by examining enzymes that form and by using the genes as templates. While not a direct measure, these profiles provide an indication of genetic diversity. By monitoring these profiles over several generations of fish, the geneticist can determine whether hatchery practices have reduced the genetic variability. If it has, corrective measures can be taken.

Genetic diversity between stocks is also important to a species. For this reason, it is forbidden to interbreed different stocks of salmon without the approval of the geneticist.

There are two general types of genetic variation: discontinuous and continuous. A trout lacking pigmentation displays a discontinuous variation that is controlled by one or a few gene loci. Most traits of economic importance, however, display continuous variation, i.e., they are controlled by many genes and the organism's interaction with the environment. Characteristics derived from continuous variation include length and weight, development time, and fecundity.

Additional information on genetics and the Department's policies on salmon stock genetics may be found in the "Provisional Policy Statement on Salmon Stock Genetics which is available from the principal geneticist (333 Raspberry Road, Anchorage, AK 99502).

Effective Population Number

You can calculate an effective population number (EPN) for establishing and maintaining genetically diverse brood stocks by using the following equation:

$$Ne = \frac{4Nm\ Nf}{Nm + Nf}$$

Where Ne is EPN
Nm is the number of parent males, and
Nf is the number of parent females.

An EPN of 400 is generally considered adequate for maintaining genetic diversity. It is the equivalent of breeding 200 females with 200 males. The greater the number, the lower the rate of inbreeding (Figure 1). "Ne" is strongly influenced by the number of the less frequent sex (Figure 2).

Note that the EPN will be equal to the actual number of spawners only when the numbers of male and female spawners are equal. The equation becomes important when fewer than 200 males or females are available for spawning. If only 150 male spawners are available, 300 female spawners would be required to achieve an acceptable EPN of 400.

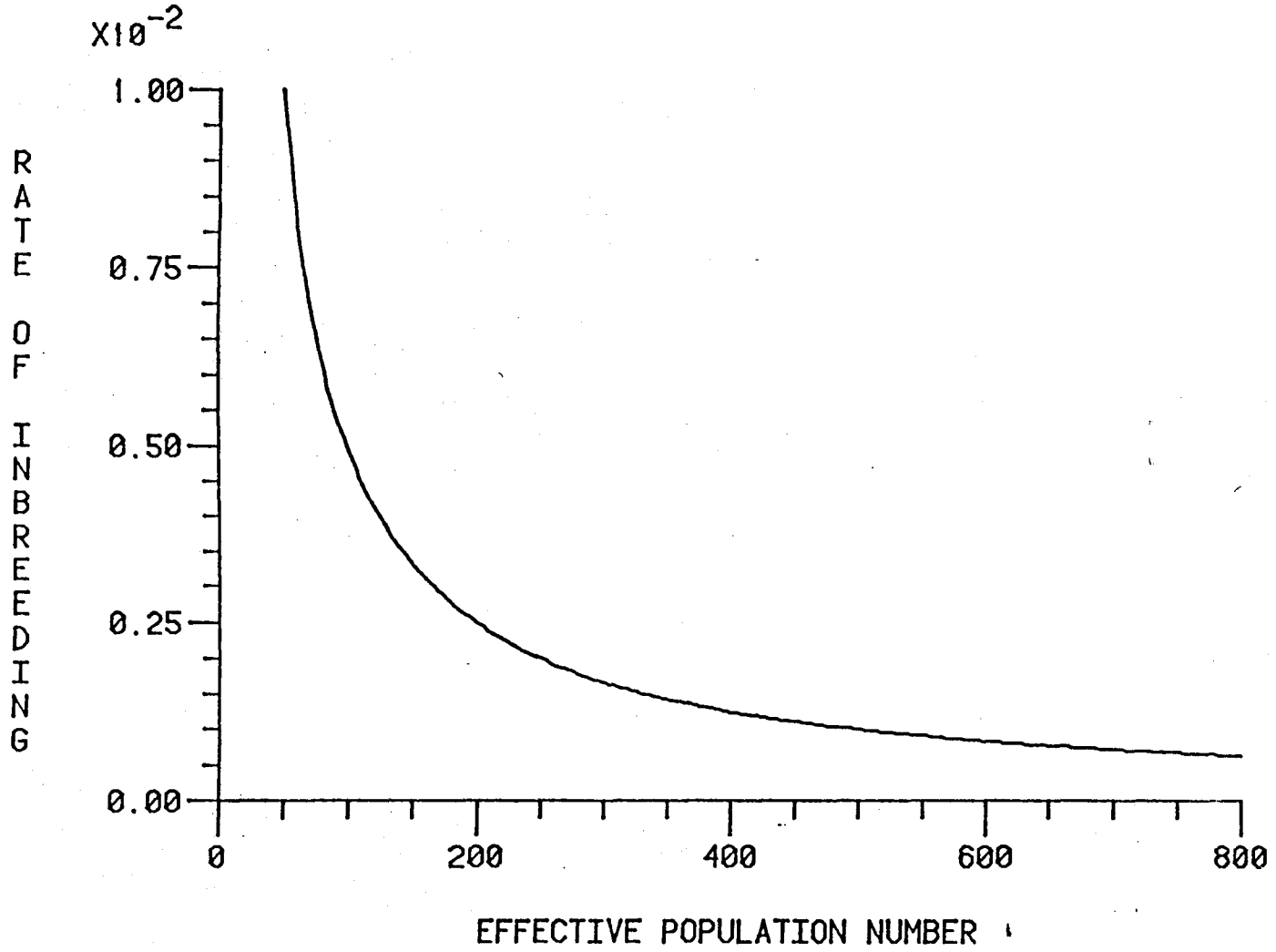

Figure 1.--Rate of inbreeding (dF = 1/2Ne) plotted against effective population number (Ne = 4NmNf/(Nm + Nf). Decrease in rate of inbreeding is small when effective population number is increased above Ne = 400. Nm and Nf are the number of males and females, respectively.

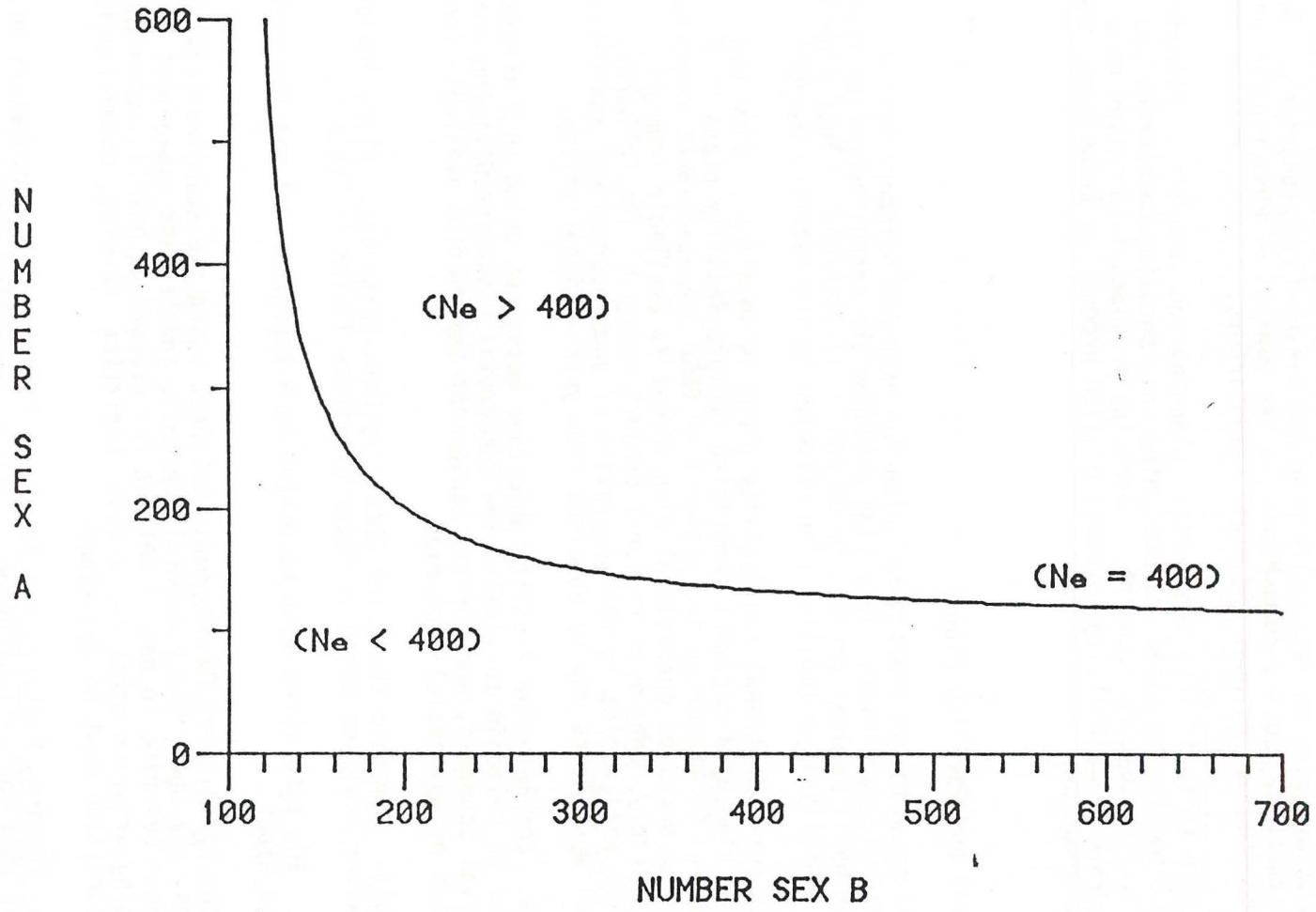

Figure 2.--Number of either sex (A) required to maintain an effective population (Ne) equal to 400 plotted against the other sex (B) which is varying. On the curve, Ne = 400. For points above and below the curve Ne > 400 and Ne < 400, respectively.

PLANNING AND REPORTING

Hatchery personnel are required to produce reports and fill out forms. The information in them is valuable for planning, designing hatcheries, and evaluating hatchery techniques. Hatchery managers themselves require detailed information to rationally plan and manage fish production. This same information, usually summarized, is the content of most reports and forms requested by regional and state headquarters.

This section is split into two parts: planning and reporting. Planning covers information required for designing and operating hatcheries and for conducting fish culture research. Reporting provides information needed for hatchery quality control, improvement of fish production techniques, and public information.

Planning

Operational and Facility Plans:

Project teams write an operational plan for each new hatchery before construction is completed. This plan outlines the annual operation from the first year until maximal production of adults is attained. This plan is modified annually in a facility plan prepared by the hatchery manager.

The format for operational and facility plans is identical. View the operational plan as a document comprising "future facility plans." A general format is presented in Chapter 7 of FRED *Organizational Management Manual*, and a detailed operational plan format is available from your regional hatchery manager or regional project manager. The following instructions will assist in the prepration of your operational and facility plans. All worksheets may be obtained from your regional office.

Objectives. Complete the Facility Objective Worksheet using FRED standard assumptions on salmonid survivals, the Commercial Fisheries Division Donor Stock Removal Schedule, and harvest management information available from the Division of Commercial Fisheries.

Water Supply. Complete the Water Chemistry Data Worksheet. Also, include complete water analyses based on water qualities listed in Table 1.

Egg Takes. Egg take sites must be marked on a topographical map for each species and stock.

Complete the Brood Stock Development Worksheet using the standard survival assumptions, the Donor Stock Removal Schedule, and harvest management information. Describe in detail methods for capturing, holding, spawning, and disposing of brood stock carcasses. Logistics, housing, communications, and transportation must be described.

Incubation, Rearing, Marking, and Release. Identify which incubators and how many of them you will be using. Include water flow requirements. Estimate green to eyed egg, eyed egg to hatch, and eyed egg to emergent fry periods for each species, stock, and lot. Describe fry emergence (volitional or non-volitional) and estimate when fry emergence will begin,

peak, and end for each species and stock. Include loadings and densities. Determine fish food requirements and complete the Feeding Schedule Worksheet. Complete the monthly Bioengineering Rearing Criteria Worksheet.

Describe marking and tagging procedures, including numbers and sizes of fish. Describe logistics, housing, personnel, and transportation at remote sites.

Describe release procedures, including the species, stock, number, and sizes of fish released. Also, mark the location of each site on a topographical map.

Summary of General Hatchery Operations. The basic operations at a hatchery not directly involved with egg takes, incubation, rearing, marking, and release of fish are described in the summary of general hatchery operations section of the Operational Plan format. In this summary, plan your monthly supply schedule. Include order and delivery dates, and the means of delivery.

List food and supplies for personnel and fuel for transportation, heat, electricity, and motorized equipment. Determine your needs for communications equipment, including mail delivery, communications for routine hatchery operations, and emergency communications.

List permanent and temporary staff required for operating the hatchery. Complete the Permanent Personnel and Temporary Personnel Worksheets. Describe weather conditions - to include the following: maximal rainfall and snowfall (monthy and annually); average rainfall and snowfall (monthly and annually); average monthly temperatures; daily record of wind speed, wind direction, and tides; seismic data; if applicable.

Describe general safety conditions at the hatchery and field sites. List first aid supplies and fire fighting equipment, including pumps. Include special pumps and pump connections that must meet or exceed State and Federal standards.

Estimate the number of visitors you will receive annually and the special facilities required, i.e., rest rooms, signs, water fountains, and show cases. Will any additional employees be needed for handling visitors?

Describe garbage disposal methods at hatchery and remote sites.

Project Proposals and Plans:

Fish cultural research and development projects require project proposals. After a proposal is approved, a project plan must be completed. Project proposal and project plan formats can be obtained from the regional biologist in your region.

Reporting

There are four types of reports that fish culturists must be familiar with:

1) Facility Development
2) Facility Operations and Production
3) Technical
4) Standard Production Data

Explanations and formats for the first two reports are provided in Chapter 10 of the FRED *Organizational Management Manual*. Technical reports follow the FRED *Style Manual*. The fourth report, a packet of forms with detailed instructions, is available at each hatchery or from your regional hatchery manager or regional project manager.

ACKNOWLEDGEMENTS

The production of this manual was very much a divisional effort. So many people worked towards its completion that the product deserves no author or editor designation and, therefore, bears neither. Those persons, listed alphabetically, who contributed are Sam Bertoni, Irv Brock, Bob Burkett, Bob Davis, Jill Follett, Dave Gaither, Roger Grischkowsky, Janet Hall, Jeff Hartman, Bernie Kepshire, Mark Kissel, Jeff Koenings, Karen LeGrand, Ken Leon, Doug Loshbaugh, Teresa McGee, Keith Pratt, Cindy Smith, Kelley Starr, and Vickie Wilson. Portions of this manual rest heavily on the work of Harry Westers.

FRED Division
Juneau, Alaska
March 24, 1983

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